SCURVY AND ITS ANAEMIA

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INTRODUCTION

Coming into contact with a case with clinical features of severe scurvy but having haematological features almost indistinguishable from severe pernicious anaemia, and yet obtaining an immediate and rapid haematological response to treatment with ascorbic acid, prompted me to consult the literature on the subject. Each new article read appeared to contradict the former so that I came away perplexed and confused. This confusion was easy to appreciate as deficiency of one vitamin usually means a deficiency in many other essential requirements. Consequently I felt that the dramatic haematological response was purely the result of supplying the necessary factor, in a case of nutritional macrocytic anaemia. This necessary factor was present in the full hospital diet given.

When I appended this conclusion to the summary of his case notes, I realised that two outstanding points were difficult to explain on this basis. Firstly, he had been admitted only two years previously, the final diagnosis on discharge being "Arthritis ?Neisserian : Anaemia". From the case notes it was easy then, to see that his "arthritis" was the pain and stiffness from juxta-articular haematomata and the spongy gums and follicular hyperkeratosis, which was recorded, completed the full picture of scurvy. The anaemia (1.79 million red cells per cu. mm.) reached only 2.96 million red cells per cu. mm. after six weeks of full hospital

diet, liver, iron, sulphonamides, vitamin B complex and ascorbic acid. A full haematological examination was, unfortunately, not performed, but it was noted that, like now, he had a histamine-fast achlorhydria. Presuming then that the cause of the anaemia was the same, I wondered why the "shotgun" therapy used then, had not produced as rapid and as complete a response as had just occurred. If this presumption was correct, the factor lacking may not have been supplied in sufficient amount. As no liver, iron, vitamin B complex or sulphonamide had been given on his second admission, the therapy common to both occasions was full hospital diet and ascorbic acid, with the latter differing in dosage and the mode of administration.

The second point I found difficulty in explaining was that his recent haematological response had occurred rather soon for a haematinic taken orally i.e. the reticulocyte peak of 21% was only 72 hours after the first mouthful of the hospital diet. From this I was prompted to believe that in some way the intravenously administered ascorbic acid (in 1,000 mgm. doses) had been responsible. On consulting case notes of other cases of scurvy admitted previously, I found that anaemias as severe had responded rapidly and completely to full hospital diet and ascorbic acid.

The final diagnosis of nutritional macrocytic anaemia on the case notes and the doubt in my mind may have remained to this day, had not two identical cases of

scurvy been admitted within a week of the others discharge. They, too, were anaemic. The opportunity seemed too good to miss so, under strict control, i.e. on exactly the same diet as they received outside - the diet on which the scurvy and the anaemia raged,-ascorbic acid only was added.

Thus started what is the main object of this thesis, i.e. :-

1. To attempt to find out whether anaemia is a feature of scurvy;

2. If so, whether it is a manifestation of vitamin C deficiency purely or the result of other accompanying deficiencies;

3. If possible to determine the mechanism of its production.

As the haematological features became so interwoven with other aspects of scurvy, which is the case with most diseases, it was found impossible not to include in this thesis other pertinent observations.

It must be stated in the beginning that no case of scurvy has been rejected as unsuitable for this thesis. The method of admission of patients to this hospital makes such selection of cases impossible. In this way it is hoped that this investigation reflects an accurate

and unbiased incidence of the various features of scurvy.

From the start, however, certain difficulties were obvious and many of these were encountered.

It would not befit the principles of medicine to withhold treatment of an easily treatable condition too long. In this respect very few indeed of these patients were unemployed, consequently many depended on their health for their daily income. Fortunately their wives and children were well cared for at their kraals from whence they had come to amass a fortune. Too long a hospital stay would defeat this object. On the other hand many were loathe to leave the hospital when the time of discharge drew near. The diet, too, became a problem. To keep a patient on a monotonous diet of mealie meal porridge, bread and black tea, despite the fact that this had satisfied his gustatory and gastronomic wants for many months, whilst the man in the next bed could eat the tasty variety of the hospital diet was to us, the executors of the programme, as trying to our sentiments as it was necessary for a rigid control. In this respect a constant watch had to be kept that no new nurse, visitor or fellow patient gave any food or drink that would upset this strict dietary control.

It was attempted to overcome these difficulties in several ways. Firstly, if possible, the patient was segregated from other patients in a single-bedded ward.

Otherwise his bed was screened at meal times. Secondly the nature of the investigation was explained, as far as possible, to the patient and this led to full co-operation on his part. Thirdly the experimental control period was made as brief as possible.

Apart from these difficulties, several - all were Bantus - spoke a language quite foreign to the usual interpreters and myself. These patients were East Africans. This difficulty had its deleterious effect on the accuracy and detail of the clinical history. To overcome this, the history was re-taken just prior to the patients' discharge as by then many could speak some English or Zulu.

Daily specimens, such as stools and urines had to be obtained. This was the most difficult of all. So often a new nurse or helpful convalescent patient would discard a valuable specimen at the crucial moment and the whole investigation would have to be started anew.

A great shortage of hospital beds was by no means a minor difficulty.

Thanks to the unfailing co-operation of the ward sister, housemen, physicians-in-charge, nurses and by no means least the patients themselves, this investigation was made possible.

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ACKNOWLEDGEMENTS

CHAPTER I.

THE HISTORY OF SCURVY.

CAPE TOWN

SEVENTEENTH

CENTURY

CAPE TOWN

TWENTIETH

CENTURY

It is somewhat of a paradox that the City which was started in 1652 to combat scurvy should three hundred years later provide material for one of the largest series of scurvy published in recent times. Any South African must thrill a little at the story of scurvy for it is intimately bound up with the story of the settlement of Southern Africa.

An unfortunate disaster started this story. In 1648 the "Haarlem", a ship of the Dutch East India Company, was wrecked on its outward voyage to the East Indies. The crew remained at the Cape for a whole season until they were picked up by the return fleet, amongst whom was one, a ship surgeon, Johan van Riebeek. Two of the shipwrecked crew, Leendert

Jansz and N. Proot submitted "A Short Exposition of the Advantages to be Derived by the Company from a Fort and Garden at the Cape of Good Hope" [LEIBBRANDT (1900)]. The photographed translation of this historic document is appended (Fig. 1).

PRECIS OF THE ARCHIVES

OF THE

CAPE OF GOOD HOPE.

LETTERS AND DOCUMENTS RECEIVED.

No. 1.—A Short Exposition of the Advantages to be Derived by the Company from a Fort and Garden at the Cape of Good Hope. 1649. 26th July.

Though some who have visited the Cape, but without paying attention to its resources, will say that the place is altogether unfit and will not repay the expenses incurred, as nothing is to be had save water and wild sorrel; and others that the Company have forts and stations in sufficient number to take care of, and therefore ought to make no more; we will endeavour to show according to our understanding, and with brevity and humility, how serviceable and necessary such a fort and garden will be for the convenience and preservation of the Company's ships and men; and also that they can be established with profit and no cost.

By making a fort and a garden adequate to the requirements of the crews of the Company's passing vessels, in the Table Valley, protecting the whole with a garrison of 60 or 70 soldiers and sailors, and likewise providing the establishment with a proper staff of experienced gardeners, a great deal of produce can be raised, as will be shown further on.

The soil is very good in the valley, and during the dry season the water can be used for irrigation as required. Everything will grow there as well as in any other part of the world, especially pumpkin, watermelon, cabbage, carrot, radish, turnip, onion, garlic, and all kinds of vegetables, as those who were wrecked in the *Haerlem* can testify.

It is also beyond doubt that all kinds of fruit trees will thrive there, as orange, lime, apple, citron, shaddock, pear, plum, cherry,

The director of the general company asked for van Riebeek's opinion on the above "remonstrances". Having remained on shore only long enough to ship the "Haarlem's" goods and crew, a period of three weeks, he felt that he could add "but little more". "If, as Leendert proposes, you order your ships to touch at the Cape, I believe that a great deal of preserved provisions would be economised on the outward voyage and likewise wine; for, if they pass without touching, they do so only for the sake of premium; the consequence is that the crews are put on short water allowance and the meat and pork are boiled in salt water. Very little fresh water is given to the crew to drink, but one or two glasses of wine are distributed to make up for it and though the wine is a cordial and strengthening, the sailors remain not the less subject to scurvy and similar diseases in consequence of the staleness of the food. But refreshed at the Cape, the voyage can, with God's blessing, be safely made to Batavia with the ordinary provisions and wine allowance and fresh water, by which the Company would be greatly benefitted, securing the health of the men, and saving a great deal of preserved provisions which are everywhere required in India, whilst now they are consumed by the crews with the least benefit to themselves" [LEIBBRANDT (1900)].

The beautiful and fertile "Table Valley" was unfortunately drought-stricken when van Riebeek arrived with his family,as leader of the settlers in April, 1652. The

enfeeblement of the men by the voyage and this absence of fresh food on arrival considerably slowed the construction of the fort. At one time fresh meat was obtained from a hippopotamus killed in the Salt River [THEAL (1907)(b)].

Progress, however, was surprisingly rapid as, in the season two years later, no fewer than twenty-one ships were revictualled with fresh vegetables on their voyage Eastward. Some of these ships had lost as many as fifty men and when they dropped anchor they had over a hundred helpless with scurvy [THEAL (1907)(d)].

The frequency of callers increased. Amongst these were two English ships. One had been at sea for eight months since leaving the port of London, with the crew almost helpless with scurvy. The very ill were hospitalised but the remainder were supplied with fresh vegetables and meat at a cost of twopence a day, per man, to the British East India Company [THEAL (1907)(c)].

A letter dated 18th October 1658 was written by J.J. Houtschip and J. Syms on board the "West Vriesland" stranded after leaving Holland five months before, with a crew of three hundred and fifty-one men [LEIBBRANDT (1899)]. "We cannot refrain from informing you of the miserable condition of the crew, of which 72 are already dead and 150 men very ill of scurvy. Though brought on shore here, we shall not be able to obtain sufficient refreshment for them, as no natives or cattle

have been met by our men sent inland for the purpose. We therefore request you to assist us with vegetables and fruit, as you may deem necessary for our condition. We sent you a letter with the Penguin, but she could not leave. We cast anchor on the 14th in Saldanha Bay, and so weak handed that we could not even furl the sails ..... ".

This gives one an idea of the straits to which they were reduced and one can credit the story of a Spanish Galleon adrift on the ocean with every man dead from scurvy [HARRIS (1938)].

An excellent description of the clinical features and treatment of scurvy is found in a report issued to the Seventeen of Amsterdam on 23rd. January 1696 [LEIBBRANDT (1896)]. "You mention that you read with great discomposure in our letter of 30th January 1695, the great number of deaths on board the outward bound ships and the equally great number of sick, and that we are to give you the reasons .....". Apart from accidents, exposure, "army" diseases, dirty and wet clothing the following appears. "..... Then there is the unvaried consumption of salt meat and pork and especially of grey and white peas which are the daily pot food, and by length of time become musty in the hold - whilst the beer likewise becomes sour. All this old pot food losing its nourishing qualities, and unable to nourish a labouring man in proportion to what he requires, labouring as he does in the heat of the day, finally weakens him so much, that he becomes sleepy and lazy, and in

the end gets the scurvy. They lose their appetite, blue nobs and blotches cover the whole body, the gums rot, the patients become shivery, are feverish and fall into fainting fits, from which dysentery results. They lose heart from want of nourishment, take to their beds and all germs of strength failing them, they die. This is the unanimous testimony of all the chief surgeons, given by order of the Governor." But this report goes on: "To prevent these diseases as much as possible, good nourishing food is required and the ships should, better than hitherto, be supplied with barley, plums, raisins and currants, which, boiled together with a good deal of mum* and now and then some Spanish wine, and given to the men morning and evening would be wholesome food."

This report was issued following the loss of men on two ships in 1693. The "Bantam" had lost 221 men from scurvy in February whilst the "Goude Buys", five and a half months out from Holland, dropped anchor at St. Helena Bay with not a dozen of the crew capable of working [THEAL (1907)(d)].

The Seventeenth Century closed, then, with the anti-scurvy settlement at the Cape in full working order, with better ship construction shortening the voyage from Europe to ninety or one hundred days but scurvy still causing terrible havoc amongst seamen.

This conception of a settlement at the Cape was not originally a Dutch one. When Vasco da Gama rounded the Cape of Good Hope in 1498, 100 of his crew of 160 perished

* - a strong sweet beer.

from scurvy [HARRIS (1938)]. This is one of the earliest reports on scurvy at sea. The earlier mariners probably escaped because they "coasted". The Portuguese used the island of Mozambique as a place of refreshment by the fleets. [THEAL (1907)(a)]. This settlement provided the requirements and shelter for the fleets should the fleets have to wait for a change of monsoon. It boasted a hospital "ready for the reception of scurvy stricken sailors and soldiers arriving from Europe".

The French, too, were having their share of scurvy at this time. In the second voyage to Newfoundland in 1535 Jacques Cartier stated, "An unknown sickness began to spread itself amongst us after the strangest sort that ever was eyther heard or seene, in so much that some did lose all their strength, and could not stand on their feete, then did their legges swel, their sinnowes shrink as black as any cole. Others also had all their skins spotted with spots of blood of a purple colour; then did it ascend up to their ankles, thighes, shoulders, arms and necks; their mouths became stincking, their gums so rotten that all the flesh did fall off even to the roots of the teeth which did almost all fall out. With such infection did this sickness spread itselfe in our three ships that we were not tenne whole so that one could help the other, a most horrible and pitiful case". - [RALLI and SHERRY (1941)]. He wintered near an Indian village near Quebec. Indians too became affected but it was noticed that one Red Indian who had been seriously ill twelve days

before, was now well. The remedy was a decoction made from the sap, leaves and bark of the "Ameda" tree. At this time 100 of his 103 men were desperately ill and twenty-five of them died. The response to the decoction was miraculous. A contemporary writer said "If all the doctors of Montpellier and Louvain had been there with all the drugs of Alexandria, they would not have done so much in a year as that tree did in six days." - [HARRIS (1938)].

From the French, too, comes another fairly distinct account of scurvy [TURNER (1911)]. This account was very much earlier, in 1260. In the memoirs of the Sieur de Joinville who accompanied the crusaders lies this description: ".... a great calamity and disease came on the army, which was of such a nature, that the flesh of our legs dried to the bone and the skin was tanned blade and earth coloured like an old boot which had been hidden for a long time behind the boxes. And in addition another trouble happened to us who had this disease, an affection of the mouth, because we had eaten these fish, the flesh of our gums rotted and consequently our mouths smelled horribly. And in the end scarcely any escaped from the disease, but all died. And the sign of death which was consequently recognised was that one commenced to bleed from the nose and then one was sure to be soon dead." Their supposed cause of this disease, "these fish", was "barbel, which is a greedy fish and always goes to dead bodies and eats them."

Yet nearly 3,000 years before this, the modern

dietary conception of the etiology of scurvy was held. If one accepts Job's affliction [Job.(40)] as being scurvy, as the first chapter of the Book of Isaiah tempts one to do, then it is of interest to note how the Lord said to Job: "Then will I also confess unto thee that thine own right hand can cure thee. Behold now behemoth, which I made with thee; he eateth grass as an ox. Lo now, his strength is in his loins ..... Surely the mountains bring him forth food, where all the beasts of the field play". SWANSON (1944) points out that the ancient Hebrews talk of meat, milk, barley and wheat as chief articles of the diet, all poor sources of vitamin C. The Jews are known to have used copper cooking vessels which would have promptly destroyed vitamin C.

MACRAE (1912) quotes Pliny in stating that even the campaigning Roman soldiers along the Rhine were not free of a disease which caused loosening of the teeth. This disease was cured by eating an indigenous plant, probably a type of sorrel. This was probably the "scurvy grass", that with watercress and oranges, was advocated by Sixteenth Century writers from the same area, e.g. Ronsseus, Claus Magnus, Echtius and Wierus [RALLI and SHERRY (1941)].

It was about this time that British reports were coming through.

Sir Richard Hawkins in 1593 recorded that 10,000 seamen had died of the scurvy in his personal experience

[HARRIS (1938)]. He wrote "The signes to know this disease in the beginning are divers, by the swelling of the gummes, by denting of the flesh of the leggs with a man's finger, the pit remayning without filling up in a good space; Others show it with their lasiness; Others, complaine of the cricke of the back, etc., all which are for the most part, certain tokens of infection. The cause of this sicknes, some attribute to sloath; some to conceite, and divers men speake diversly ..... That which I have seene most fruitful for this sicknesse, is sower Oranges and Lemmons, and a water which amongst others (for my particular provision) I carryed to the Sea, called Doctor Stevens, his water .....".

The story swings back to the arrival of Lancaster's fleet in Table Bay in 1600 on their way to India. All his ships were riddled with scurvy except one. On this ship each sailor had been given lemon juice daily [LAIDLER (1936), RALLI and SHERRY (1941)]. At the Cape they, as did the Dutch ships that followed later, ate scurvy grass, fresh water and meat for which they bargained with the Hottentots. ".....as soon as they taste the shore, eat three leaved grass and fresh meat and the like and bathe and frolic in a small space." [LAIDLER (1936)].

It is surprising indeed that this experimental proof by Lancaster in 1600 did not convince all, that scurvy was easily prevented by a substance which could be carried in a condensed form. Only two hundred years later was it made a

regulation in the British Navy [HARRIS (1938)]. This step was due to the insistence of Sir Gilbert Blane, who acted upon the work of James Lind and Captain Cook.

In 1747 Lind performed his momentous experimental work on twelve scorbutics on board the "Salisbury" at sea. Given a diet common to all, "Two of these were ordered each a quart of cyder a day. Two others took twenty five drops of elixir vitriol, three times a day upon an empty stomach; having their gruels and their other foods well acidulated with it, as also the gargle for their mouths. Two of the worst patients, with tendons under the ham rigid (a symptom none of the rest had) were put under a course of sea water. Of this they drank half a pint every day, and sometimes more or less, as it operated by way of gentle physic. Two others each had two oranges and one lemon given them every day. These they ate with greediness, at different times, upon an empty stomach. They continued but six days upon this course, having consumed the quantity that could be spared. The two remaining patients took the bigness of a nutmeg three times a day of an electary recommended by a hospital-surgeon, made of garlic, mustard-seed, rad. raphan, balsam of Peru, and gum myrrh; using for common drink barley water well acidulated with tamarinds; by a decoction of which with the addition of cremor tartar, they were greatly purged three or four times during the course. The consequence was, that the most sudden and visible good effects were perceived from the use of oranges and lemons; one of those who had taken them being at the end of six days fit for

duty..... The other was the best recovered of any in his condition; and being now deemed pretty well, was appointed nurse to the rest of the sick.

Some persons cannot be brought to believe that a disease so fatal and dreadful can be prevented or cured by such easy means. They would have more faith in some elaborate composition dignified by the title of an antiscorbutic golden elixir or the like ..... Facts are sufficient to convince the unprejudiced ..... It is no easy matter to root out old prejudices or overturn opinions which have acquired an establishment by time, custom and great authorities."

Captain Cook sent an account of his experiences on his voyage round the world to the Royal Society in 1776. He was forthwith elected a member and his paper honoured with the prize medal. Apart from "rob of lemon and orange", he advocated sour krout as it "spoils not by keeping". [HARRIS (1938)].

Prior to this BACHSTROM recorded in 1734 how a sailor in the Greenland ships was so bad from scurvy that his comrades put him ashore leaving him to perish. He could only crawl but "grazing like a beast of the field" he plucked up with his teeth a plant covering the ground. In a short while he was perfectly recovered. [HARRIS (1938)].

It would be incorrect to attribute the progress in scurvy in the Eighteenth Century to the British only.

BAUMANN (1939) states how Kramer in 1720, an Austrian Physician was faced with a severe epidemic in a field army in Hungary. In answer to his appeals a shipment of dried antiscorbutic herbs was sent to him but failed to cure the disease with the result that thousands died. He wrote: "Scurvy is a terrible disease for which there is no known cure. Medication does not help, neither does surgery. Be careful of bleeding, shun Mercury as a poison. The gums may be massaged, the stiff joints may be rubbed with fat, but all in vain. If one could have at hand oranges, limes and lemons, or their preserved pulp or juice so that lemonade could be made out of them, or administered as such in 3 - 4 oz. doses. Then one could be in a position to cure this disease without other help."

Meanwhile at the Cape, although the incidence of the disease was dropping, the Dutch were still losing ships due to scurvy stricken crews. In 1747 the "Reyersdal" ran aground and immediately broke up between Dassen and Robben Islands with only twenty-five men surviving. She was four and a half months out from Holland with 125 men dead from scurvy whilst those of the remaining 83 weakened diseased crew, were too few to manage the ship. [THEAL (1907)]. In 1778 the "Venus" from Batavia just reached Delagoa Bay with a loose rudder and only eighteen men able to man the ship.

The "Snoek" left Flushing on the 10th June 1768 and reached the Cape of Good Hope on the 17th November, 1768, with 30 dead and 58 sick, "mostly down with scurvy".[STAVORINUS (1798)].

At the turn of the Eighteenth Century the Cape changed hands, and it was the British's turn to report on scurvy from this area. Rear-Admiral Pringle in 1797 requested reinforcements to garrison the Cape as "the few people to be procured here, being little more than equal to the Deaths and other Casualties. I am sorry to add that the Scurvy has made very considerable progress in the ships returned from sea.... ". Whereas in 1802, about the time of compulsory lemon juice in the British Navy, H.M.S. "Tremendous", fourteen weeks from Bombay, docked in Simon's Bay with her crew greatly afflicted with scurvy. All, except one, were cured with a liberal supply of "fruit and vegetables, with fresh meat and other comforts". [THEAL (1898)].

The confusion of the word lemon and lime not only led to the nickname "limies" but also, by virtue of the poor antiscorbutic properties of preserved lime juice, led to the doubt as to dietary origin of scurvy. This doubt persisted throughout the Nineteenth Century and into the beginning of this Century.

Consequently the regulation issue of lemon juice in the British Navy, was not adopted by the Merchant Navy until a Parliamentary enquiry was held in 1865. This enquiry showed how ship owners and ship masters were sceptical of the value of lime juice. The reason was soon made clear because of twenty-five samples of lime juice from merchant vessels, analysis showed that eleven were diluted, twelve were solutions

of citric acid, some of which contained sulphuric and others acetic acid. [TURNER (1911)]. Despite this, however, lime juice was made compulsory on the merchant ships.

It is no wonder then that the Potassium theory, citric acid theory, and acidosis theory [RALLI and SHERRY (1941)] reigned supreme each in their turn, until just over thirty years ago, due in all probability to the inconstant effect of lime juice.

An excellent example of this is found in a report from Brown at the Poles only as recently as 1920 [RALLI and SHERRY (1941)]. He stressed that it was not the climate but scurvy that gave the polar regions their bad name. At Cape Flora the leader and staff lived chiefly on fresh bear meat, the crew stayed on board consuming tinned meat, tinned vegetables and the compulsory lime juice. The crew developed scurvy. A similar story emerged from Franz Joseph Land in 1894-95 where the crew on board the "Windward" had lime juice daily and all developed scurvy [TURNER (1911)].

Scurvy was still playing havoc on land. During the Eighteenth and Nineteenth Centuries, scurvy, if not in military campaigns, generally occurred in prisons, insane asylums, poor houses or houses of refuge and correction [RALLI and SHERRY (1941)]. Scurvy was rife in the Crimean War where 23,000 cases broke out in the French army alone [WATKINS-PITCHFORD (1912)]. Likewise it took its toll in the American

Civil War, the first World War and the Russo-Japanese War. After the siege of Port Arthur, about half of the garrison of 17,000 had scurvy [RALLI and SHERRY (1941)].

At the Cape, however, reports from inland did not mention the disease. Lichtenstein travelled inland from 1803 to 1806 and devoted two chapters of his book to disease [LICHTENSTEIN (1930)]. No mention of scurvy appears in either Whites or Blacks. HOFMEYR (1941), however mentions that from the pioneering missionaries occasional reports of the existence of scurvy amongst the natives were received.

This statement is not confirmed when the returns of Hospital admissions in various areas of the Cape are studied. The "port" was Cape Town where at the Somerset Hospital in the years 1855 to 1865, there are records of 135 cases of scurvy, four of whom died. Whereas at Port Elizabeth Hospital - not a frequently used port then - only five patients were admitted from 1859 to 1866 and at Albany Hospital, one hundred miles inland, this diagnosis was not made on any case admitted. - [HOSPITALS, ASYLUMS and HEALTH 1856 - 1880].

Possibly with the Great Trek, the Boer with his superb marksmanship kept himself well supplied with fresh meat. He also planted fruit and vegetables wherever he and his family settled.

In the Boer War, however, many reports of scurvy are to hand [MACRAE (1912), SPENCER (1912), GRAHAM (1912) and

TURNER (1911)]. Turner describes how previous to this, scurvy was prevalent in the jails and prisons but in 1897 and 1898 it was particularly troublesome. He attributes this to the condition of the country at the time. There was a native war (the Langberg campaign), rinderpest had carried off nearly all the cattle and a long continued drought carried off the remainder. A serious shortage of labour occurred at the Kimberley diamond mines. Here the native labourers were living on kiln-dried imported meal. Nearby, at Klipdam, a special scurvy hospital for 400 to 800 patients had to be run by the Government. Spencer likewise felt that the outbreak in the Middelburg jail was due to imported meal. Good fresh meal caused an improvement. MACRAE (1912) shows how the Bantu was free of scurvy whilst he followed the British troops and ate their rations, but once he had to fend for himself scurvy was common. European and Bantu prisoners in Gaberones, Bechuanaland, received the same mealie meal diet but the Europeans received meat in addition. Scurvy broke out among the Bantus and was cured by adding fresh meat to their ration. With improvement in the prison diet scurvy has almost been banished from South African Gaols. In many places they now serve a role of "nutritional hospitalisation" as the hungry Bantu, raw from the country, is often driven to petty crime thereby.

"Those who know the kaffir and his haphazard habits in regard to securing food" [MACRAE (1912)] will infer that scurvy depended on the goodness of his surroundings. The

devastation of crop and cattle during the Boer War left the Bantu in a sorry plight.

In the 1914 - 1918 War the Bantu suffered again but here scurvy broke out three to four months after their arrival in France from South Africa [DYKE (1918)]. Dyke pointed out that scurvy often was seen in the Spring in South Africa after the previous crop of kaffir corn had failed. He attributed this outbreak to the use of Kaffir Beer made in France from ungerminated millet which is poor in vitamin C. He suggested good Kaffir Beer made from germinated millet.

Later DELF (1922) showed the antiscorbutic properties of Kaffir Beer in the monkey.

In the meantime Axel Holst and Frolich in 1907 had produced scurvy experimentally in the guinea pig and the epic race for the discovery of this antiscorbutic vitamin started. Zilva in London, King, Vedder and Lawson in America, Bezssonoff in France with Tillmans and Hirsh of Germany and the Hungarian chemist Szent-Gyorgyi working at Cambridge and later in Hungary, were the competitors. [HARRIS (1938)], [RALLI and SHERRY (1941)]. The final discovery and synthesis probably resulted from Zilva's and Tillmans's use of the redox dye (2:6 dichlorophenolindophenol) to distinguish fresh fruit juices with high antiscorbutic value from the stale juices having poor or no antiscorbutic properties. It is hard to believe that in a short space of twenty years the cause and

final solution to a disease that had shaped the course of History from Biblical times, was discovered. No longer does the dread of scurvy hang over the progress of civilisation. With education of the people into correct dietetic habits and the easy transport of synthetic vitamin C for long expeditions or military campaigns, scurvy is fast dying out in civilised areas.

The war against scurvy was by no means entering its final stages in South Africa, however. A large epidemic amongst the Bantu mine labourers in Wankie, Rhodesia in 1909, was reported by HEWETSON (1910) who felt that dietary factors played no part in the etiology of scurvy. The magnitude of the problem of scurvy on the Rhodesian and Witwatersrand mines led to the appointment of a commission, the findings of which were published in 1910 [FLEMING et al (1910)]. In a report which could hardly be improved upon to-day they explain the factors underlying the prevalence of scurvy in Rhodesia. "....1,671 cases were reported in Rhodesia in 1908 with 207 deaths, equivalent to a rate of 13.5%, but the Commission is convinced that the reported cases by no means represent the total number occurring." The rarity of scurvy at the kraals led to the conclusion that the "wants of an idle kraal native are not to be compared with those of a mine labourer". They were unanimous in their opinion that scurvy would be eradicated with improvement in the diet and strongly recommended Kaffir Beer. It was, furthermore, their feeling that the recruits needed a long rest

on a liberal diet as often they arrived after a long and arduous journey, fed merely mealie meal and salt en route, "with the disease upon them or develope it within three months."

An excellent account of his experiences with scurvy on the Kimberley mines and elsewhere was published a short while later by TURNER (1911). He discusses fully the compound system, where the labourers are housed in living quarters but usually have to provide and cook their own food, a scheme still in existence in some areas to-day.

HOFMEYR (1941) delves further into this problem in his review on the history of vitamin C deficiency in South Africa. The impact of civilisation and the expansion of modern industry has led to the transfer of large numbers of aborigines from a natural life that they and their ancestors have understood for centuries, to living and feeding conditions approaching that of the white man but completely foreign to them.

Almost ironically came the report of an epidemic in Rhodesia by DRY (1933) in the same year as the sensational synthesis of vitamin C.

Antiscorbutic campaigners in South Africa were not dormant, however. The problem, thanks to the advance of chemical methods in determining the value of various antiscorbutic foods, [LEVY and FOX (1935)], was slowly being solved. Scurvy, on the mines, was slowly disappearing. Supervision of cooking and of the rations issued was probably responsible.

Suddenly in 1939, scurvy again raised its ugly head. This led to an intensive investigation by FOX et al (1940) where the same problems of the Rhodesian Commission in 1910 were brought to the fore. Here, as before, it was the recruit arriving with low vitamin C stores as a result of his dietetic habits which included excessive cooking, the use of iron pots, and the aversion to vegetables and to fresh milk.

The problem remains the same in certain areas to-day and with the rising cost of antiscorbutic foods and with the increasing exodus of the raw uneducated, untrained Bantu to the larger towns, it may become even worse.

The approach to the problem is made even more difficult by the mentality of the type of patient liable to scurvy and by the mentality of the Bantu himself. Divorced from his leisurely home life the rural Bantu seeks employment in positions where only unskilled, usually strenuous labour is available to his capabilities. Continuing on his precarious level of vitamin C intake with its consequent low store, he stands or falls in his adaptation to a life quite foreign to him.

It is true that with education and urbanisation the Bantu is learning the European mode of life. His country cousin is still the same however. Life-long habits are hard to change, especially in a superstitious mind. He still consumes the same porridge and kaffir beer that he has consumed for centuries. Neither he nor his ancestors have eaten the green

vegetable or orange on which we set so much store, and manifest scurvy is almost unknown in the kraal or rural life of the primitive Bantu. It is not difficult, then, to understand his outlook when he laughs at the suggestion that such an incapacitating disease is caused merely by dietary mismanagement. Neither is it difficult to understand that the problems faced thirty years ago still remain the problems of to-day with regard to the rural Bantu as he, as an individual, has changed in neither outlook nor habits. How aptly do Lind's words of just over two hundred years ago, quoted on Page 12, apply to the Bantu of to-day.

Civilisation is rapidly advancing, however. Ignorance is being eradicated in the correct way i.e. from where it starts in the kraal. Dotted throughout the country and native reserves are Government Health centres. The Bantu is being trained to cultivate his land, to grow vegetables, to get used to eating them, and to cook them properly. Every year some advance, however small, is made. Every year more children and more mothers are being educated into a new way of life. Education of the grown-up raw Bantu male is, however, no easy matter, as with any primitive superstitious people. The rising incidence noted in the following chapter reflects well the magnitude of this problem; a problem made no easier by the rising cost of living.

It will be seen, then, that there is still a long road to travel before a state comparable to that of more

civilised countries is reached; a state where reports of scurvy are appearing less and less and where the presence of a predisposing factor and not merely gross ignorance has to be postulated for the occasional occurrence of a case. The final history of scurvy in South Africa, then, has yet to be written.

Milestones in the history of scurvy

B.C. : Biblical reference suggestive that part of Job's affliction was due to scurvy.

I260 : Sieur de Joinville's account of scurvy among the Crusaders.

I498 : Vasco da Gama's voyage round the "Cape of Storms", I60 men - I00 dead from scurvy.

I520 : Magellan's crew riddled with "The Plague of the Sea and the Spoyle of Mariners".

I535 : Jaques Cartier's men cured of scurvy with decoction from spruce tree.

I593 : Sir Richard Hawkin's : Treatise on scurvy.

I600 : Captain Lancaster arrived at the Cape without a case of scurvy on board his ship due to a daily ration of lemon juice.

I648 : Wreck of the "Haarlem" at the Cape.

I649 : Rescued crew of "Haarlem" suggest refreshment station at the Cape.

I652 : First settlement at the Cape.

I696 : Report to Amsterdam on the causes of the numerous deaths at sea and probable causes of scurvy.

I747 : James Lind : Experiment in the cause and treatment of scurvy.

I776 : Captain Cook's account of scurvy acclaimed by the Royal Society.

I795 : Lemon juice issued as a daily ration in British Navy.

I795 : Cape taken over by the British.

I798 : Stavorinus :"Voyages to the East Indies".

I848 : Garrod's potassium theory of the cause of scurvy.

I854 : Compulsory issue of Lime juice in British Merchant Navy.

I865 : British Parliamentary enquiry into scurvy on the ships.

I894 : Scurvy among crew aboard "Windward" in Antarctic, in spite of daily lime juice.

I896 : Regulation re compulsory lime juice on ships re-inforced.

I899 : Citric acid theory (Netter).

I900 : Boer War : Scurvy among refugees in Concentration Camps.

I902 : Scurvy among S.African Bantus in post-war period.

I906 : Reign of the acidosis theory of scurvy. (Sir Almroth Wright).

I907 : Holst and Frolich produced scurvy in guinea pigs.

I909 : Epidemic among Bantu mine labourers at Wankie (Southern Rhodesia).

I909 : Commission of Enquiry into pneumonia and scurvy among mine labourers in Rhodesia and the Transvaal.

I9I8 : Great War : Scurvy among S.A. Bantus in France.

I922 : Delf : Antiscorbutic properties of "Kaffir Beer".

I924-I930 : The increasing use of the "redox " dye for assessing the antiscorbutic properties of various foodstuffs.

I933 : Isolation and synthesis of vitamin C.

CHAPTER II.

ETIOLOGY OF SCURVY.

A. Introduction.

THE MATERIAL OF THIS STUDY.

Thirty-two adult patients admitted to the medical wards of Groote Schuur Hospital, Cape Town, with the diagnosis of Scurvy, from January 1945 to August 1950, formed the basis of this study. Six additional patients admitted with the same diagnosis are not included as no accompanying haematological data were available. Thirteen of the thirty-two patients have been under my direct observation (Cases 1 - 13) and are the main subjects of this study. Cases 14 to 32 were studied only through data obtained from the hospital records, and personal discussion with those in whose charge these patients were treated.

The completeness of these records in the earlier years (1945 to 1947) depended largely upon the competence of the house physicians, and it is quite possible that minor features unrecognised to the inexperienced eye, yet noticed subsequently by the more senior and experienced physicians were not recorded. With the increase in staff at this hospital over the last three years this possibility has largely been eliminated. Fortunately for this survey the majority of these admissions took place during this latter period and it is indeed pleasing to see the detail and completeness of these latter case-notes. However, for the sake of statistical accuracy in sections on the incidence of certain features and

detailed haematological progress, reference in these sections will be made to which cases are being discussed - e.g. either Case 1 to 13 designated the "Personal" series or the total series of Cases 1 to 32.

Apart from the routine history taking and physical examination conducted in all the cases, the "Personal" histories (Cases 1 to 13) were re-taken when the patients were well. This was found to be a most useful manoeuvre as by the time of their discharge many patients could speak English or Zulu, making interpretation unnecessary. Although this did allow more accuracy, the Bantu is not usually a good witness. Despite the many long hours spent in taking detailed medical and dietary histories, it is with respect to their duration particularly, that the above statement must be borne in mind.

Each case studied was a consecutive admission to the medical wards. No case has been rejected on the basis of it not being "suitable" for this thesis. In this way an attempt has been made to obtain as far as possible a true or accurate incidence of certain features of scurvy. The Bantu, however, does not feel disposed to consulting medical opinion unless he is incapacitated by his disease. Consequently almost every case was severe.

In order to determine the specificity of certain features, it was necessary to be sure that they responded to pure synthetic ascorbic acid only. Consequently eleven of the thirteen patients in the "Personal" series were placed on the

same diet on which they developed the disease, as soon as they were admitted. Variable control periods to observe the effect of rest in bed alone, or of other synthetic substances, were instituted. By virtue of the fact that these patients depended on their work for their financial well-being, these control periods could not be too long. Each patient had full insight into the nature of the investigation and was fully co-operative.

B. Occurrence in This Series.

1. AGE.

The patients ranged in age from 22 years to 60 years with the majority being between 20 and 40 years (Fig. 2). This then is a considerably younger series than the usual series, large and small, of more civilised races, and areas where scurvy appears, in most cases, to be confined to males over 50 years of age.

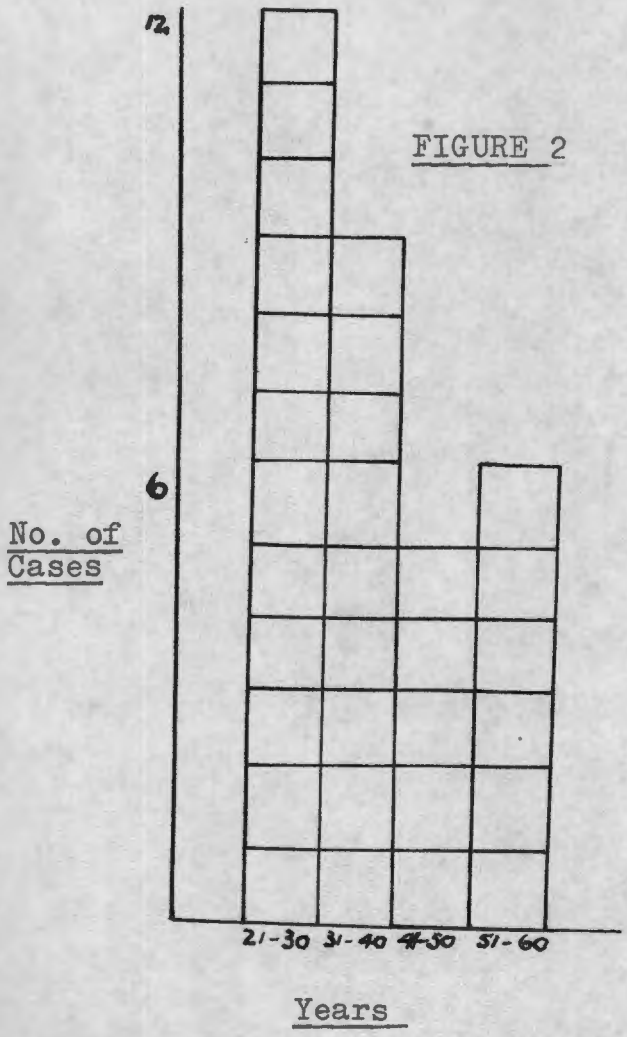


FIGURE 2

2. SEX.

All cases were males.

3. RACE.

All cases, except one, were Bantus. The exception was a Cape Coloured aged 52 years (Case 23). Seventeen of the Bantus were East African Natives (Nyasas), seven were Xosas, two Pondos, one Basuto and one Zulu, while three with no tribal denomination given, completed the tribal incidence.

4. SEASONAL.

Of the thirty-two patients studied one was admitted in

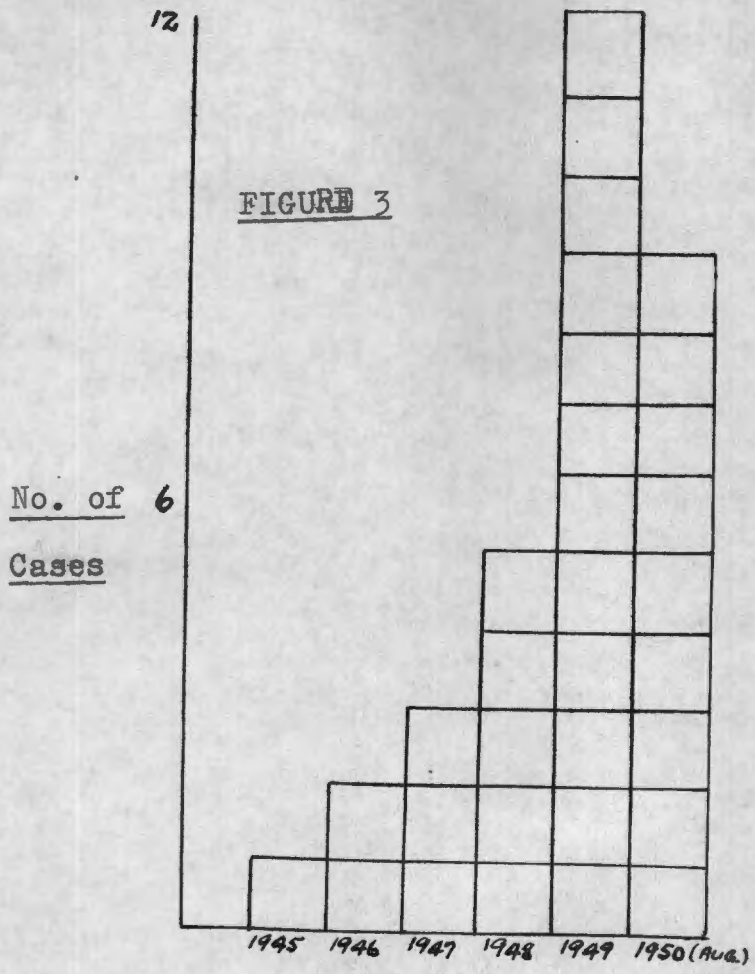


FIGURE 3

1945, two in 1946, three in 1947, five in 1948, twelve in 1949 and nine during the first eight months of 1950 (Fig. 3). The majority were admitted in the Autumn and early Spring (Fig. 4).

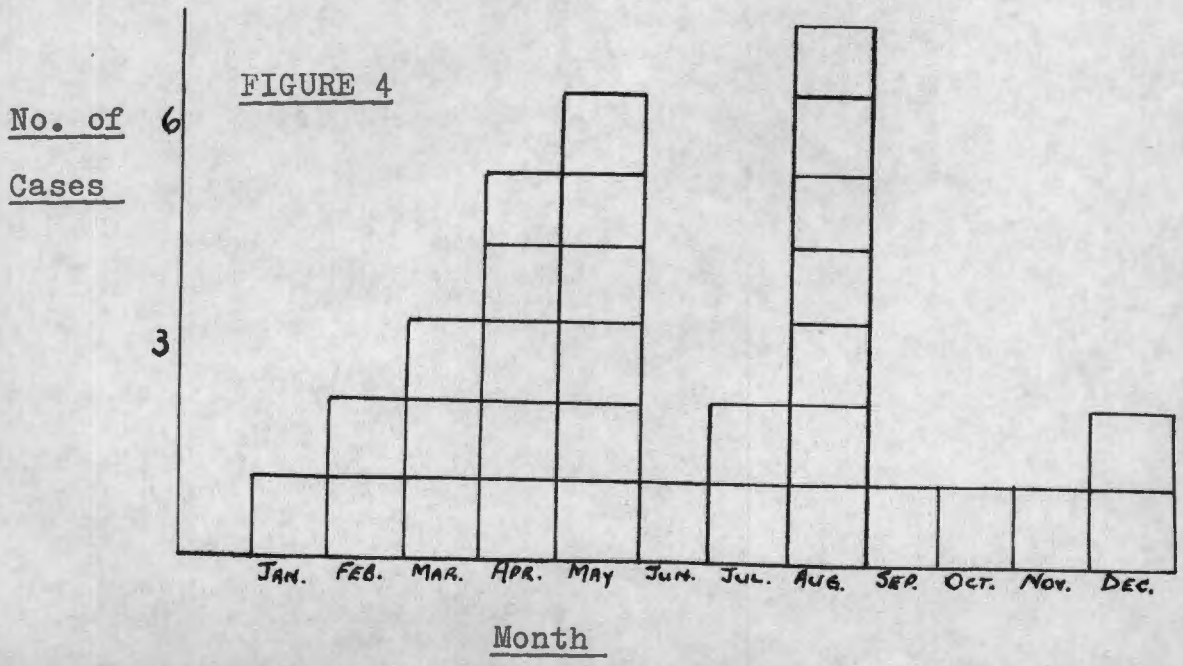


FIGURE 4

C. Etiology.

1. EXCITING FACTOR.

No doubt rests in any mind to-day that scurvy results from a dietary deficiency. The essential factor is vitamin C which has now been synthesised in a pure state and called "ascorbic acid". Although this disease has ravaged the world for centuries, hung like a cloud over every early seafaring ship and shadowed campaigning armies, the tremendous advance in our knowledge of scurvy has only occurred in the last twenty or thirty years. It is, indeed, a tribute to the earlier workers that scurvy is now considered a rare disease, being now sporadic in more civilised areas, yet still endemic in the Bantu but rarely if ever epidemic.

In the decade prior to the discovery of ascorbic acid in 1932 [HARRIS (1938)] the trend of medical feeling had swung back to the views of the early navigators that scurvy was a dietary disease. The floundering in the depths of uncertainty of previous workers must have been painfully obvious to patient and doctor alike. Controversy raged as to the efficacy of potassium citrate against lime juice, citric acid etc., whereas in the meantime the patient was responding to fresh food. Controversy also raged over its contagious possibility, a possibility due to its epidemic nature with some isolating micro-organisms and others not. Deficiency of potassium salts, lack of vegetable acids such as citrates and lactates were as vehemently favoured as acid intoxication

eventuating in a defect of blood coagulation or the theory that it was a type of ptomaine poisoning from eating tainted meat [TURNER (1912), WATKINS-PITCHFORD (1912), MACRAE (1912)].

Even after the discovery of vitamin C some [CROFT and SNORF (1939), CRANDON et al (1940), LOZNER (1941)] wondered whether the full blown syndrome of scurvy was really due to deficiency of ascorbic acid alone.

All cases in this series gave a dietary history of a gross lack of vitamin C. The content of the diet was remarkably common to almost all and almost identical with the diets recorded by the early South African writers, mentioned above.

The basis was a thick, dry porridge made of coarsely ground mealie meal, cooked for a long period in iron pots over an open fire. This was eaten three times a day. To this bread was usually added but fish, meat or small dry brown beans were a luxury. Black tea or coffee completed the meal. Sour milk occasionally was placed on the porridge. The few who had eaten vegetables had done so very infrequently and had chopped them up into small portions and boiled them over prolonged periods in an open pot with their meat as a stew. The residual water and fluid was usually discarded. Only one had consumed "kaffir beer" - a brew made from the sprouts of kaffir corn - but that had been in small quantities, a month prior to admission. No fruit of any kind had been eaten over the preceding year.

2. PREDISPOSING CAUSES.

This particular diet is, however, consumed by thousands of Bantus throughout the country, even in the same areas or compounds from which these patients came, and it differs little from the diet consumed in their kraals, yet scurvy is not invariable. Furthermore, it is well-known that vitamin C containing foods may be in the ration yet scurvy still results. The existence of predisposing causes has thus been postulated.

The deficiency of an essential nutrient is dependent on an inadequate supply for the body's needs which may be absolute or relative, the latter being due to increased utilisation of that factor or increased loss from the tissues. Absolute deficiency may be due to the consumption of foods lacking in vitamin C, destruction of the vitamin in overheating, bruising or in using excessive alkalis in the preparation and cooking of the food, apart from allowing the food to become stale [FOX (1940)]. Even should all these factors be guarded against, there is still the possibility that a high gastric pH may destroy the vitamin before it even reaches the small intestine, where a defect in absorption may further limit its entrance to the tissues. Diarrhoea may play a part in the latter mechanism.

Man, like other primates, guinea pigs and certain ungulates cannot synthesise vitamin C, therefore he is wholly dependent for his supply on food sources. Considerable con-

troversy exists as to his daily requirement, however. One finds that competitors in the last pre-war Olympic games chose a diet containing 300 mg. vitamin C daily. [FOX (1940), BROCK (1942)]. RALLI and SHERRY (1941) recommend 100 mg. a day, SMITH (1938) 50 to 60 mg. daily while the League of Nations Technical Commission (1938) recommended only 30 mg. a day. This confusion probably results from different standards having been adopted. Three possible levels of requirement may be considered:-

(a) the indispensable minimum required to prevent the onset of scurvy,

(b) the adequate to cover the requirements for normal adults with their own inherent variability with various activities and environments,

(c) the "saturated" level or luxus consumption level - dependent on biochemical tests.

ZILVA (1944) based his conclusions on deducing the human requirements from that of the guinea pig - the deduction [KELLIE and ZILVA (1939) with WOLBACH (1937)] being a fair degree of probability. Two mg. ascorbic acid daily not only prevents the production of macroscopic and microscopic lesions of scurvy in a guinea pig on a scorbutogenic diet, but apparently also enables the growing animal to lead a normal existence. About ten times this dose is required to attain "saturation". In man "saturation" requirement is, as

measured by the urinary output of ascorbic acid, of the order of 50 mg. a day. Using the ratio of the guinea pig, the minimum human requirement would be in the region of 5 mg. per day - a surprisingly low figure. The Medical Research Council [PETERS et al (1948)] took up the challenge and on a series of experiments on twenty human volunteers came to the conclusion that the "minimal protective dose" as measured by the criteria of the presence of scurvy was "in the region of, perhaps somewhat below 10 mg. daily". To allow a margin of safety the requirement was finally fixed at 30 mg. daily.

Although this is probably the most accurate assessment of the daily requirement, it merely confirmed a finding of 350 years ago. One may recall the first expedition of the newly chartered East India Company in 1600 when Queen Elizabeth laid the foundations of the British Empire in India [RALLI and SHERRY (1941)]. Months later when the small fleet of four ships reached Table Bay, Captain Lancaster of the Dragon, after bringing his own ship to anchor, had to get out his boats and send men aboard each of his consorts to do the like for them as their crews were too weakened by scurvy to make the effort. The healthy condition of the Dragon's men was due to the fact that their farsighted commander had taken along a supply of lemon juice and given three spoonfuls to each sailor daily i.e. equivalent to approximately 7.5 mg. daily.

This small daily requirement then would adequately explain, as FOX (1940) points out, the long periods for which healthy individuals can subsist on experimental extremely

deficient diets, and the rarity with which scurvy appears amongst the communities who are compelled to do the same. The occasional consumption of a vitamin C containing food may avert the danger of scurvy which may be perilously near.

This occasional intake of ascorbic acid makes an accurate assessment of the incubation period in clinical scurvy impossible. Furthermore the type of patient liable to develop scurvy is not a good witness. Together with this the incubation period must vary with the adequacy of the previous diet and consequently the stores in the body, and the presence or absence of the predisposing factors to be mentioned.

RALLI and SHERRY (1941) quote George Whetstone who in one of the first works on tropical medicine in 1598 said of scurvy: "that it is so ordinary at sea as it hath been seldom seene, any ship or pinnace to be foure months upon any voyage ..... ". In the Precis of the Cape Archives LEIBBRANDT (1896) one may see a letter written by two unfortunates on board the "West Vriesland" on 18th October, 1658, in Saldanha Bay. "We left the Vlie on the 18th May with 351 men, very few of whom at present are well; yea! so few that we are at present unable to weigh anchor to go up a little higher into the bay". Thus it appears that in the old days the incubation period varied from 100 to 150 days. Indeed it is fortunate for Cape Town that the sailing ships did take so long, otherwise it might never have been founded.

It is surprising then that in more recent times under the ideal condition of young men on diets adequate in every respect but for vitamin C [CRANDON et al (1940), PETERS et al (1948)], the incubation period was approximately the same, i.e. 120 to 150 days. A much shorter time was reported by ANDREW (1949).

In this series language difficulty and the type of patient made an accurate assessment of the incubation period impossible. The shortest duration of a diet devoid of vitamin C was stated to be three months and the longest two years with average falling between five and seven months. Probably the most accurate was Case 10 who had been discharged "saturated" with vitamin C four months prior to the onset of his first symptom.

Thus when an inadequate amount of ascorbic acid in a diet exists, a gradual insidious onset of scurvy will follow within three to six months. This long incubation period is in keeping with the small daily requirement postulated.

It is conceivable, however, that the road to scurvy may be shortened by other factors. Increased demand by the body for ascorbic acid by using up the available stores may hasten the onset. At the same time this mechanism may apply when an adequate vitamin C content of the diet exists and cause a relative vitamin C deficiency.

Infections have been blamed as the most potent cause of this by many authors [METTIER et al (1930), DRY (1933), UNGLEY (1938), FOX (1940), EDDY and DALLDORF (1941), KEKWICK et al (1947) and SHAFAR (1949)] but the question whether it is cause or effect arises. Certainly both in guinea pigs and infants with scurvy, infection frequently supervenes. In man, CRANDON et al (1940) and PETERS et al (1948) showed that during experimental human scurvy no increased incidence of infection occurred, except that the latter noticed an exacerbation of acneiform papules that had been present before the experiment started. Crandon stated that he was never more free from upper respiratory infection than during his scorbutic period. No drop in complement occurred. RALLI and SHERRY (1941) mention, however, that the large haematomata may undergo abscess formation.

On the other hand EDDY and DALLDORF (1941) felt that many manifestations of Typhoid fever, the Zenker's degeneration and intestinal bleeding, were due to superimposed vitamin C deficiency, a view which is not now held. METTIER et al (1930) report the appearance of scurvy after an attack of Bronchitis in one case.

With regard to the Bantu, DRY (1933) and KEKWICK et al (1947) mention intestinal parasite infestation and malaria as probable predisposing causes. Shafar mentions that the acute stage of malaria increases the requirement of vitamin C.

It was at first thought that this would explain the higher racial incidence in East Africans.as malaria is endemic in this region. In nine of these eighteen a hard firm splenomegaly was found but in the three in the "Personal" series with the same, no signs of activity could be detected. Only in one case (Case 10) was intestinal infestation found. This was a tape worm (Taenia saginata).

The presence of a slight dry cough in so many of the cases in the "Personal" series caused routine X-Ray chest examinations. SHAFAR (1949) mentions that tuberculosis in the active state is associated with heightened demands for ascorbic acid. In only one case - the case with the tapeworm - was there an infra-clavicular lesion and this appeared inactive. Several sputum examinations failed to reveal acid-fast bacilli. In the remaining nineteen cases of the total series two had suggestive evidence of tuberculosis. This is discussed in greater detail later. The cough mentioned above in the other cases frequently disappeared even before vitamin C therapy was given. No other signs or history of previous infection were elicited. It was felt that infection played little, if any, part as a predisposing cause in this series.

Hyperthyroidism, pregnancy, rapid growth and muscular exertion have been mentioned as increasing the requirements of vitamin C. In this respect it is of interest to note that CRANDON et al (1940) noted a progressive drop in the basal metabolic rate. This was considered as insignificant

as the drop before was the same as after the beginning of vitamin C administration.

That scurvy improves with bed rest alone, even on a vitamin C free diet, is well known [SHULTZER (1936), UNGLEY (1938), FOX (1940)]. A similar state of affairs occurs in other deficiency diseases, e.g. Pellagra [VILTER et al (1945) SHAFAR (1949) and BARKER (1950)]. In fact SHULTZER (1936) noted how scorbutics put to bed showed gradual improvement in the clinical features but on allowing the patients up, these features recurred and after three to four days of intravenous therapy with ascorbic acid these features rapidly subsided.

That the converse holds true, however, i.e. that increased muscular exertion hastens the scorbutic progress, is, according to the literature, a controversial point.

Stress does appear to modify the site and extent of the lesions. In olden days blacksmiths and woodcarders had their lesions mainly in the shoulders and arms, whereas in soldiers they were mainly in the calves [FOX (1940)].

CRANDON (1940) noticed that immediately after a fatigue test, while in the scorbutic state, he showed for the first and only time a small haemorrhage at one gingival margin, a finding that suggested to him that had he been exposed to such fatigue throughout the experiment he might have developed signs of scurvy much earlier. DRY (1933) mentions that the probable reason for the rarity of scurvy in the Bantu in his native

environment and its ready occurrence when he leaves to work in the more civilised areas is the question of muscular exertion. In all the cases of this series the subjects performed fairly strenuous physical work, e.g. concrete mixers, quarry workers, carriers, etc.

This view was not held by the very early writers on scurvy. Lind remarks that the "indolent and slothful sailor" was more often stricken with scurvy than the one who was active - [FOX (1940)]. Of course, the former unfortunate sailor might have been scorbutic at the time of his "indolence".

FOX et al (1940)(1941) in their survey of scurvy on the Rand gold mines are of the opinion that the degree of muscular exertion has no effect on the development of the disease. The incidence amongst those doing heavy manual work was no higher than amongst those doing light work. Some workers have noticed a decrease in the urinary excretion of vitamin C during exertion but BERNSTEIN (1937) obtained similar urinary excretions for Native mine labourers whether they were working or resting. Furthermore a sudden onset of the disease is the exception and usually cases develop months after arrival at the mines.

BERNSTEIN (1937) felt that this increased utilisation could be due to increased sweating and loss of vitamin C via this mechanism. Experimentally he showed that this loss could be as great as two mg. per hour which agreed well with

CORNBLEET et al's figures (1936). The urinary vitamin C content remained unchanged before and after this experiment. WRIGHT and MacLENATHEN (1939) disagree in that the loss is much lower and the loss during sweating is due to increased utilisation in the tissues, they state. Further work is necessary to clarify these problems. It is of interest here that all the cases in this series were in occupations where sweating, undoubtedly, is fairly profuse. Furthermore the seasonal incidence is at its highest at the end of the hot Summer. That loss via the sweat mechanism may be the reason for increased utilisation of vitamin C in acute malaria, other infections and hyperthyroidism, is not mentioned.

A low renal threshold [UNGLEY (1938)] has been postulated as a possible mode of increased loss of the vitamin. CRANDON et al (1940) believe that the threshold drops in the totally deficient state and rises again as "saturation" with vitamin C takes place. FAULKNER and TAYLOR (1938) feel that ascorbic acid is a threshold substance with a critical level of excretion in the vicinity of 1.4 mgm. per 100 ml. serum. KELLI and ZILVA (1939), however, maintain that no constant renal threshold exists for vitamin C but that there is competition for the ascorbic acid of the blood by the absorptive capacity of the tissues and the excretory function of the kidney. FOX (1940) suggests that in prolonged ascorbic acid deficiency compensation, in preserving the body stores, takes place by a reduced glomerular filtration. FRIEDMAN, SHERRY, RALLI and

RUBIN (1940)(1938) showed that even at the lowest plasma levels the reabsorption of the vitamin is never complete and there is a minimum amount excreted in the urine. Evidence appears to disfavour a low renal threshold as a predisposing cause of scurvy.

In this respect arterio-sclerosis has been mentioned by the earlier writers [MINOT (1929), METTIER et al (1930)] as a predisposing cause but this is due more likely to its frequent occurrence in elderly solitary men.

Yet another mechanism may operate in causing increased loss of ascorbic acid from the body. The vitamin C content of the stools which is usually a constant amount normally, becomes markedly increased in diarrhoeic states [SHAFAR (1949)]. Case 9 in this series gave a history of diarrhoea just prior to the onset of his symptoms. Intestinal hurry may, however, prevent adequate absorption taking place.

Intestinal hurry and very low sugar tolerance curves are features associated with the "deficiency pattern" of the small bowel on barium meal examination, described by GOLDEN (1941). It is a matter of dispute whether this pattern is a cause or effect of certain deficiency syndromes with which it has been associated. In scurvy UNGLEY (1938) without further comment, felt that disorders of motility and absorption may be predisposing factors. It is of interest to note that in over 30% of the East African Bantus studied by KEKWICK et al (1947) a "deficiency" small bowel pattern was present. Due

to this he compared the effects of the intravenous against the oral "saturation" test but found no significant difference with regard to absorption in these cases. Large doses were, however, used. Consequently therefore it would be difficult to apply the same conclusions when only a border-line intake is present. That failure of absorption is a feature occurring during the development or early stages of scurvy in man is refuted by CRANDON et al (1940). No alteration in the glucose tolerance curve occurred.

In only two of the eight cases examined in this series, was a deficiency small bowel pattern found. In neither of these cases had any vitamin C containing foodstuffs been consumed over the previous year. Although final and conclusive evidence is lacking it was felt that as a mechanism this played a very minor part, if any, in this series.

Evidence exists, however, that in spite of adequate amounts of ascorbic acid in the diet some factor may operate in preventing vitamin C from reaching the tissues. This evidence is based on observations made by earlier writers. HON (1935) noted that ascorbic acid was twice as effective in guinea-pigs when given hypodermically than orally. In man SHULTZER (1933) held the same view. DRY (1933) used orange juice intravenously in preference to the oral route. WRIGHT and LILIENFIELD (1936) even induced relapses by placing several scorbutics on oral vitamin C and cured them again by giving it intravenously.

These facts would suggest then, that some factor was preventing the absorption of ascorbic acid. Ascorbic acid, however, is a very unstable substance chemically and it is conceivable that it may be destroyed before it is presented to the small intestine for absorption.

For many years a comparison has been drawn between the metabolism of iron and the metabolism of ascorbic acid. MOORE et al (1939) in their work on iron metabolism showed that the effect of gastric acidity was to delay the formation of insoluble compounds until they are brought into contact with the reducing forces of the small intestine. Although they obtained satisfactory rises in the serum iron levels after iron ingestion in achlorhydria they warn, however, that these results may not apply to the effect of anacidity on small amounts such as in the food. They felt that with the large doses used some iron almost certainly reached the small intestine before neutralisation took place.

Applying this to scurvy one may see that in achlorhydrics on a high daily intake scurvy need not occur as some may get through to the small intestine. In those who are on a border-line intake, however, the presence of achlorhydria may be a serious matter. Scurvy has been cured by doses as little as 20 mg. orally daily [BARNES (1947)], 40 mg. orally [DUNLOP and SCARBOROUGH (1935)] and 50 mg. orally [VAUGHAN (1934)]. Unfortunately the presence or absence of gastric acid is not stated. Other authors who have reported achlorhydria have given

large doses, i.e. 500 to 700 mg., by mouth. The lowest recorded dose in a definite achlorhydric scorbutic which produced a response appears to be Case 8 in this series. He received 100 mg. daily orally. However, the point at issue is whether an intake level of the neighbourhood of 10 mg., the minimum protective dose, is affected or not.

Exact proof then is lacking but indirectly many points in this respect are of interest.

It is well known that ascorbic acid is unstable in an alkaline medium. Peptic ulcer therapy aims at producing an alkaline medium. Furthermore most peptic ulcer diets consist of milk, eggs, bread and fish - all low in vitamin C content. Plasma levels, which reflect the daily intake, have been found to be low in these cases [BOURNE (1938), PORTNOY and WILKINSON (1938), LAZARUS (1939) and CROFT and SNORF (1939)]. However manifest scurvy although described [DAVIDSON (1938)] is rare. LUDDEN et al (1941) showed that no interference with vitamin C absorption was observed during the administration of alkalis. These low levels may then merely reflect the low intake in the diet.

ALT et al (1939) found low plasma levels in pernicious anaemia and iron deficiency anaemia with achlorhydria. This work was not confirmed by CAYER et al (1946) in pernicious anaemia. However an anaemia which responded to ascorbic acid, in pernicious anaemia cases, occurred during the food shortage

in the early War period in Great Britain [DYKE et al (1942)].

It must also be shown whether achlorhydria, which is common in scurvy [MINOT (1929)], produces the disease or is a result of the disease, either directly or via the associated anaemia. In the latter case the achlorhydria would then be an unspecific manifestation of anaemia only - experimental proof being shown by APPERLY and CARY (1936).

Many authors have noted the common association between scurvy and achlorhydria [SHULTZER (1933), JENNINGS and GLAZEBROOK (1938), McMILLAN and INGLIS (1944) and VILTER et al (1946)]. All but one of these cases were anaemic however.

It is of interest to note that in experimental human scurvy on a diet adequate in all respects bar vitamin C, CRANDON et al (1940) noted the presence of hypochlorhydria and after vitamin C only was added to the diet the response to histamine was much higher.

It is not clear whether the subject had hypochlorhydria prior to the experiment or not. This is of importance in that many people without apparent disease may have achlorhydria and that this incidence increases with age [VANZANT et al (1932), BLOOMFIELD and POLLAND (1933), LANDER and MACLAGAN (1934) and DOIG et al (1950). In the Bantu this incidence appears to be much higher as the rarity of peptic ulcer would suggest [BARNES and GORDON (1937)].

Table 1.

Case No.	Age (Yrs.)	Before Vitamin C			After Vitamin C		
		% P.C.V.	Before Histamine	After Histamine	% P.C.V.	Before Histamine	After Histamine
1	30	14	0	0	36	0	0
2	37	37	0	0	40	0	0
3	40	20	0	0	42	0	0
4	49	23	0	17			
5	25	22	0	0	40	0	14
6	28	15	0	0	41	0	0
7	50	53	6	40	53	8	32
8	35	22	0	0	41	0	0
9	24	23	0	0	42	6	13
10	60	26	7	20	41	62	78
11	27	42	10	not given	45	30	43
13	59	15	15	11			

Gastric Analyses : Represents highest unit of free acid. All cases except Case 1 had vitamin C only added to the control diet. The analysis was repeated between three and five weeks after treatment. Each case had pepsin present in normal quantities.

Consequently to show that achlorhydria is a feature of scurvy is just as difficult as to show that it was instrumental in the production of scurvy.

In the"personal" series, fractional test meals were performed on twelve patients (see Table I.). Provision has been made in the table for comparision with the age and packed cell volume at the time of the test. One other case in the total series (Case 27) with histamine-fast achlorhydria and severe anaemia is not included in the table.

It will be seen that eight of these thirteen cases showed a histamine-fast achlorhydria. In all but one a severe anaemia was also present. Seven of these eight were repeated not less than three weeks after vitamin C therapy commenced. At this time there was no associated anaemia. Five of these seven still had histamine-fast achlorhydria, one showed free acid only after histamine and the seventh had 6 units free acid before and thirteen umits after histamine. In every case pepsin was present in normal quantities, the average pH being in the region of 3.8 to 4.2. In the other cases repeat meals showed that in the non-anaemic scorbutic (Case 7) no change occurred with vitamin C, whilst in the other with a normal packed cell volume (Case II), an increase was found. Thus in four of the ten cases followed up, a response was shown to vitamin therapy but in only one of these, could this response be attributed to the vitamin C alone and not the concomitant improvement in the associated anaemia. Does the persistence of the achlorhydria after vitamin C therapy in 60% of the cases studied mean that it

/may

have contributed towards the production of the scurvy, as certainly from the meagre data available, achlorhydria as a feature of scurvy is not proven.

It has already been mentioned that the part played by achlorhydria must apply only to low intake groups, otherwise one would expect relapses soon after discharge. Only two cases were re-admitted in the whole series and one of these had free acid. The part played by achlorhydria then must be a very minor one and further work is necessary to clarify the possibility that ascorbic acid is destroyed by the lack of acid in the stomach.

It is well known that faulty preparation of foodstuffs may lead to the destruction of ascorbic acid before they are consumed. Mention has previously been made how, the ability of fresh orange juice to reduce the redox dye as opposed to inability of synthetic or juice that was not fresh, to do the same, played an important part in the discovery of vitamin C. The early mariners knew that fresh vegetables were essential. Ascorbic acid is unstable not only in prolonged exposure from storage prior to cooking, but after cooking also. As FOX (1940) points out, bruising, rough handling or mincing damages the cell walls causing the liberation of destructive enzymes; contact with some metals e.g. iron and particularly copper hastens the destructive process. Prolonged heating, especially in the presence of alkalis as in the making of a stew, is very destructive, especially if the cooking water, into

which most of the ascorbic acid is extracted, is discarded.

UNGLEY (1938) aptly sums up by stating that scurvy is common in men "left to the mercy of their own culinary incompetence".

The untrained Bantu male is neither a dietician nor a cook. He unwittingly pays no heed to each one of these precautions. He cooks his food in uncovered iron pots over open fires. He prefers his mealie meal porridge cooked until it is of a much drier consistency than that preferred by the European. If he felt inclined to buy vegetables he would come away from the outlying store with a cabbage that drank the fruits of the earth long before. Even should it be fresh he is not gentle by nature and he is economical in his travels. When that cabbage eventually approached the pot it would have been chopped by his rough hand and blunt knife into small pieces to commence the prolonged simmer into a stew with the meat until the latter was cooked. The residual water to him is no delicacy but helps to clean out his pot.

The Bantu, however, is leisurely. To him food is necessary merely to satiate his appetite. Mealie meal porridge provides this bulk, needs little skill at preparation and is not expensive. Vegetables, on the other hand, are expensive and the rising cost of living may explain the increasing number of admissions over recent years. Another reason may be in the increasing exodus of the rural Bantu to town. This exodus is for a specific purpose. He comes to

town to make his fortune, but ill-equipeed with the means to do so. Being an unskilled labourer and unfamiliar with the European languages he is precluded from such positions, e.g. domestic servants, waiters etc., where properly cooked and well balanced diets are provided for him. Instead he is engaged in fairly strenuous manual labour in mixing concrete, working in quarries, etc. He is given shelter in the neighbouring compound where, without instruction, he is thrown to the mercy of his own dietetic ignorance. In himself, however, he is content. His diet satisfies both his stomach and his pocket. Three quarters of his salary is weekly added to his rapidly growing fortune. The quicker this is done, the sooner may he return to his leisurely life at the kraals. Achievement of purpose, without sacrifice, is an accomplishment of which few can boast. The Bantu achieves his purpose but, unwittingly, his sacrifice is his health.

Slowly scurvy descends upon him. The painful gums and loss of appetite create the vicious circle in decreasing the intake still further. His well-being is preserved, however, and not until the incapacitating haematomata appear, does he find difficulty in holding his job.

So constantly did this type of story emerge that three or four compounds in the neighbourhood provided 90% of the material studied.

For example the detailed history of one patient

ran as follows. Three years before he was in Nyasaland. He was quite healthy until a few months work at a fish factory. Prior to this he had worked on some mines for a smaller salary but had quarters and food, cooked for him, provided free. This was not good enough. At the fish factory the employees received up to £4. a week. Now he had to provide and cook his own food. At first his appetite was satisfied by mealie meal porridge, meat, bread and coffee.Potatoes and vegetables were far too expensive, he stated. Soon he found that two loaves of bread a day were quite sufficient and much cheaper. Meat, now, was an occasional luxury. Just before admission even one loaf of bread was painfully enough for his lessened appetite and tender gums. Nevertheless he was quite proud of the fact that he was achieving his purpose in sending more than three of his four pounds a week home.

In only two cases could poverty be blamed for the deficient diet. No urbanised Bantu appeared in this series. In all cases the basic diet, as described, was the same. Similar too was purpose for thrift. In both married and unmarried it was to buy cattle, the index of wealth of the rural Bantu. To the unmarried the cattle are necessary for "lobola" - the marriage fee demanded by the prospective father-in-law.

A visit to one of these compounds would show many Bantus existing under identical conditions in identical surroundings, eating the same type of food, cooked the same way, yet only a small percentage develop scurvy. Reasons for this

individual susceptibility have been advanced already but no convincing solution to this problem has emerged. Nor was this solution to be found in KEKWICK et al's (1947) suggestion that other vitamin deficiencies were responsible. In fact one was impressed with the surprisingly well nourished appearance and the surprising lack of other vitamin deficiencies in this series.

There was a marked difference in racial incidence in this series with the East African Natives (Nyasa) being far more commonly affected than the South African Natives (Xosas etc.) It was along these lines that it was hoped the solution to the individual susceptibility, in this series, lay.

It seemed peculiar that the patients from one compound were East Africans (Nyasas), while most of the labourers there were South African Bantus (Xosas, etc.). The only difference in their mode of life was that the Nyasas would not partake of the Xosa "Kaffir Beer" - a beverage of low alcoholic and low vitamin C content (about 0.8 mg. per 100 ml. [FOX and STONE (1938)]) but consumed in large quantities.

This simple explanation of the racial incidence would not explain the completely opposite state of affairs on the Rand Gold Mines where Cape or South African Natives (Xosas, etc.) develop scurvy far more frequently than East Africans [FOX and DANGERFIELD (1940)].

The natural habitat of the two races differs. The uninitiated traveller in the South African Native Reserves may

be surprised at the striking absence of any green vegetable or fruit cultivation. With Health Centre development these conditions must improve but at the onset the Xosa and his kind are suspicious and usually somewhat averse to eating the green vegetables of the white man, a change of living that may well interfere with his leisurely disposition. Soil erosion from overgrazing, cattle tracks, broadcast sowing and the ignorance of contour ploughing makes conditions less attractive in educating the Bantu into a new way of living. Fresh cow's milk is also not to their liking, despite the fact that the index of wealth is the number of cattle owned.

Despite these conditions, together with the usual droughts, as a visit to the Polela Health Centre in the heart of a Natal Native reserve showed, manifest scurvy is almost unknown [MAFUTO (1950)]. This has been noted by others [DRY (1933), BARKER (1950)]. The rural Bantu is a man of leisure. His womenfolk and offspring hoe the mealie fields and even brew his Kaffir Beer and evidently from the sprouts of the kaffir corn enough vitamin C is obtained to prevent scurvy. This Kaffir Corn is used either in the making of this Kaffir Beer or ground up as a meal, by crushing between stones in the more primitive areas, or by machinery owned by the trading storekeepers. This meal is then made into a porridge (mabela porridge) and forms the basis of the diet. Variety in the porridge line is attained from grinding his mealie as well. In some areas wheat is grown. This is taken to the storekeepers and on the percentage mill depends its colour and vitamin B content. The Bantu

prefers taking his wheat to the storekeeper who gives him the "whitest" mill! All varieties of dried beans, except green beans, are grown and eaten. Meat is a luxury but their cattle provide their source of greatest gustatory pleasure - sour milk.

It is not then a diet that would build up a large reserve of vitamin C in the body. Added to this are the effects of droughts, poor soil and overcooking. TURNER (1912) explains the absence of scurvy in the working Bantu women and children as due to a more varied diet. In the fields they find roots and catch rats and mice which are eaten fresh.

Other factors may however operate. EDDY and DALLDORF (1941) present evidence of the probable presence of adjustment or compensation after prolonged vitamin C lack. The guinea pig's organ vitamin C stores fall and then rise slowly so that they are greater at the end of the first month than at the fifteenth day. Furthermore symptoms could be retarded or minimized by withholding water. They assume that this may be due to a reduced rate of glomerular filtration. This is of interest as the patients with scurvy in this series showed a decreasing urinary output as the disease progressed.

In East Africa, however, home conditions appear to differ. Here paw-paws, cabbages, tomatoes, guavas and oranges grow freely, and the Nyasas questioned in this series stated that they consumed liberal quantities of these foodstuffs at home. Unlike the Xosas they were fond of green vegetables.

This, however, is not confirmed by KEKWICK et al (1947) who studied ascorbic acid metabolism of East African Bantu soldiers on a calculated intake of 15 mg. per day. Their plasma levels compared well with FOX's (1940) low levels in the South African Bantu. On the assumption, however, that these levels could be raised with the addition of further ascorbic acid to the diet they felt that they represented a deficiency and were not physiologically low normal levels. In analysing the discarded portions of an orange ration, Kekwick et al showed the East African's dislike for oranges, especially those of the sour variety, which resulted in only about 30% being consumed. It is noteworthy that this daily intake of 15 mg. is well above the minimal protective dose, which would explain their statement that cases with manifest scurvy were very uncommon.

The contradictory racial incidences of Fox et al's survey (1940) and this series may be due to these tribal dietetic habits. On the gold mines an adequate supply of vegetables is available. The Xosa discards this and gets scurvy. Where however the position is reversed, i.e. where the Bantu has to buy his own food, and this means an adequate supply of vegetables does not exist to either the Nyasa or Xosa, it is the Xosa who survives. This must be due either to the process of adaptation from an almost lifelong suboptimal vitamin C intake or, as mentioned before, to the consumption of some other vitamin C foodstuff such as Kaffir Beer. Scientific proof is necessary to disprove the fact that the latter mechanism is the more likely.

It will be seen then that factors operative in this series are akin only in some ways to the predisposing factors mentioned in reports from more civilised areas. The frequency with which single and solitary men were affected was noted by SHULTZER (1933), UNGLEY (1938) and others. The position is well summed up by McMILLAN and INGLIS (1944) as: (a) Ignorance of the necessary food factors, (b) Apathy as they required preparation, and (c) Poverty, making it impossible to buy an adequate diet or reside in lodgings with proper cooking facilities.

A small precentage develop in food fads, the disabled or mentally defective, and those on special diets such as ulcer diets [DAVIDSON (1938), METTIER et al (1930) and LAZARUS (1937)].

It is the conclusion of this survey that in this series absolute deficiency or lack of vitamin C containing foodstuffs due to ignorance, ignorance of these essential food factors and ignorance of their proper preparation, was the outstanding factor in the production of scurvy. Once scurvy with its painful gums was present, a vicious circle began to operate, decreasing this intake still further. Other predisposing causes such as achlorhydria, defective absorption, diarrhoea, loss via the sweat and increased metabolic demand, must play their part only when the intake is low, and are of secondary importance.

It is felt that individual susceptibility in this

series is best explained, again, on this concept of absolute deficiency, i.e. a matter of degree. The occasional consumption of some vitamin C containing foodstuff such as Kaffir Beer, was enough to avert disaster in those not affected.

SUMMARY OF THE ETIOLOGY OF SCURVY.

1. The etiological aspects of scurvy have been discussed with respect to a series of thirty-two cases of scurvy here presented.

2. The majority of these cases were Bantu males under 50 years.

3. The largest number of admissions occurred at the end of the Summer and it is suggested that extra loss of vitamin C in the sweat, tipped the balance against them.

4. An increase in the number of admissions has occurred over the latter years either due to the increased exodus of rural Bantus to the towns or the rising cost of living.

5. In all cases there was sufficient cause for scurvy in the grossly deficient diet, although this diet apparently did not produce other vitamin deficiencies.

6. The higher incidence in the Nyasas than in the South African natives was discussed with regard to their home conditions and usual dietetic habits. It was felt that the consumption of Kaffir Beer was the most probable cause of this different incidence.

7. To explain individual susceptibility, possible predisposing causes, other than ignorance of essential food factors and their method of preparation, were discussed. These included achlorhydria, defective intestinal absorption, diarrhoea, increased utilisation such as in infections and hard muscular

effort, and increased loss as with diarrhoea, a low renal threshold and in the sweat.

8. That the minimal daily requirement is very small - in the region of 10 mg. daily - was noted.

9. Due to this the incubation period of scurvy is long - in the region of one hundred days.

CHAPTER III.

THE SYNDROME OF ADULT SCURVY.

The Clinical Features, with other factors that may complicate the syndrome.

THE CLINICAL FEATURES OF ADULT SCURVY.

RALLI and SHERRY (1941) sum up their long review on scurvy by stating "As one looks back at the reports of the Seventeenth and Eighteenth Centuries it is significant that with scurvy as with many other diseases the earlier clinical descriptions of the disease have not been improved upon." More exact knowledge as to the specificity of certain features, by their response to orange juice and later pure ascorbic acid, has been made possible by the experimental production of scurvy in the guinea-pig and later in man. This has further helped to answer the question as to what part, if any, factors such as multiple avitaminosis and infection may have played in the syndrome of scurvy and its complications.

As shown in the guinea-pig [EDDY and DALLDORF (1941)] and by autopsy studies in man [GORDON (1942)], the underlying mechanism of the syndrome is haemorrhage throughout the tissues, particularly in the subcutaneous tissues, deeper fascial planes and intermuscular septa. In the lower limbs there may be large extravasations of blood between the muscle groups.

It is to the magnificent work on human volunteers that one may turn to obtain the most accurate and earliest features of scurvy [CRANDON et al (1940), PETERS et al (1948)]. All are unanimous that scurvy has a gradual and insidious onset, the first feature being

1. Skin Changes.

The nature of the skin change in every case was

uniform. It was:-

(a) Enlargement and keratosis of the hair follicle.

ANDREW (1949) noticed its appearance after about twelve weeks and that after adding only ascorbic acid to his diet, it gradually disappeared over the following month and no trace of the condition remained after two months. No volunteer in the report by PETERS et al (1948) failed to develop follicular hyperkeratosis on a diet adequate in all respects but vitamin C. They first noticed it after seventeen weeks of vitamin C deprivation, six of the ten volunteers had the feature at twenty-one weeks and all ten had it by twenty-six weeks. The appearance and disappearance of this feature strictly reflected the intake of ascorbic acid only. Consequently they proved beyond doubt that these were the earliest features of scurvy. Giving only 10 mg. ascorbic acid daily, by mouth, the skin appeared normal after seven to nine weeks. CRANDON et al (1940) noted the appearance of hyperkeratosis follicularis after nineteen weeks of a diet adequate in all respects but vitamin C.

Even in 1919 the significance of this lesion was noted by WILTSHIRE (1919) who provided a detailed description of its appearance clinically and this accurate description was confirmed by skin microscopy thirty years later, with very few new features added [PETERS et al (1948)]. Horny epithelial debris accumulates at the follicle mouth, plugging it and giving rise to a hard conical swelling, a pin's head in size. Later this becomes flattened - often appearing as a comedone - with a hair curled up inside. If this hyperkeratotic plug is picked

off, a small slightly bleeding crater would be left [CRANDON et al (1940)].

The distribution of this skin change follows a definite pattern, e.g. arms and forearms, particularly the extensor surface; the extensor and flexor surfaces of the thighs, calves and shins; the back and buttock particularly at sites of pressure, e.g. belt or elastic; rarely the abdomen and chest, never the face or inner aspect of thighs [WILTSHIRE (1919), CRANDON et al (1940), PETERS et al (1948)].

It was for this reason that the series presented in this thesis was divided into two groups. In the "Personal" series of thirteen cases this sign was particularly sought for and in every case it was found (See Table 2). Here it was most commonly found on the extensor aspects of the thighs, particularly the lower half and on the posterior aspect of the forearm in its upper half. In each case the hyperkeratotic follicles were as described above and in those cases where the plug was scraped off, a tiny slightly haemorrhagic crater remained. The hair would then uncurl and stand erect in its follicle with the horny plug flying as a flag attached to its distal end. (See Figs. 5 and 6). Owing to the black skin it was often easier to appreciate the hyperkeratotic follicle by palpation. It is for these reasons that remarks were made in the introduction to Chapter II regarding the fact that it was quite possible that minor features, unrecognisable as significant to the inexperienced eye, were not noted in the

particular case's notes.

FIGURE 5

Follicular hyperkeratosis on anterior aspect of the lower third of the thighs.

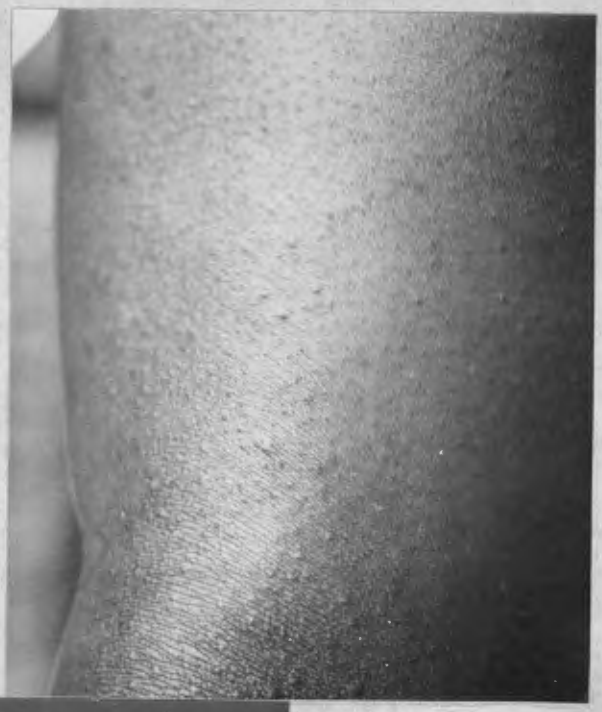


FIGURE 6

(elbow)

As stressed by many [FOX (1941), PETERS et al (1948)] more attention should be paid to this simple yet significant clinical sign, especially in deciding on a gum lesion. It has never failed to appear as the first sign in experimental human scurvy [CRANDON et al (1940), PETERS et al (1948), ANDREW (1949)] and in no case in the "Personal" series was it absent although in one or two it was present only on a small area above the knee or on the ulnar border of the forearm near the elbow.

It is with some surprise that on consulting

certain textbooks both on general medicine and the avitaminoses [CONYBEARE (1940), EDDY and DALLDORF (1941), EDDY (1941) and HARRIS (1938)], no mention is made of this as a scorbutic sign. Furthermore perusal of the larger series of case reports on scurvy in the current literature [McMILLAN and INGLIS (1944), VILTER (1946)] showed that in those cases where it was mentioned, it was included under "features of multiple avitaminosis", i.e. under vitamin A deficiency.

Reports of night blindness in scurvy by earlier workers and follicular hyperkeratosis in scurvy has brought forth comment on the similarity between lesions of vitamin A and vitamin C deficiency [SHAPIRO (1942)]. No case in this series complained of night blindness and reports of this as a feature of scurvy are not now seen. The specificity of follicular hyperkeratosis in scurvy has been proved beyond doubt. When Crandon developed this feature his plasma vitamin A level and dark adaptation test was quite normal. Furthermore he was receiving a vitamin A supplement at the time. In this respect it is of interest that STANNUS (1945) maintains that follicular hyperkeratosis has nothing to do with vitamin A deficiency, and experimentally induced vitamin A deficiency in man appears to confirm this [GILDER (1950)].

It is also of some interest with regard to the question of whether such a state as subclinical scurvy exists that the plasma vitamin C was zero ninety [CRANDON et al (1940)] to one hundred days [PETERS et al (1948)] before this sign appeared and that most patients are unaware of its existence

[WILTSHIRE (1919)]. Furthermore with regard to subclinical scurvy only in one case [CRANDON et al (1940)] were there any subjective or objective features of ill-health, prior to or during this period. In this one case there was impaired ability to perform aerobic work but no impairment of anaerobic work.

(b) Perifollicular Petechiae.

By use of the skin microscope it was shown that after a few weeks the enlarged follicles turned red due to congestion and proliferation of the blood-vessels round the hair follicles [PETERS et al (1948)]. This gradually increased and within another week or two the enlarged hair follicles became haemorrhagic, the red colour turning dark purple and no longer disappearing on compression. With this microscope many red cells could be seen outside the vessels. Small perifollicular haemorrhages or petechiae were noticed by Crandon about a month after the appearance of the hyperkeratosis. They were confined to the lower legs and occurred in greatest number after he had been standing for some considerable time at an operation. RALLI and SHERRY (1941) remark that petechiae, mainly at the site of hair follicles, occur where the capillary pressure is high such as in the legs or in regions distal to constricting bands of clothing. They were found in fifteen of their twenty-three cases whereas VILTER et al (1946) found them in sixteen out of their nineteen cases. McMILLAN and INGLIS (1944) record them in all their fifty-three cases, being the only sign in two of them. DRY (1933) also comments on their presence in the Bantu and like KEKWICK et al (1947) feels that they cannot be detected

easily in black skins. The same difficulty was noted in this series. In the lighter skinned Bantus it was however easily perceptible (see Figs. 7 and 8), the latter showing as a ring of darker pigmentation around each hair follicle

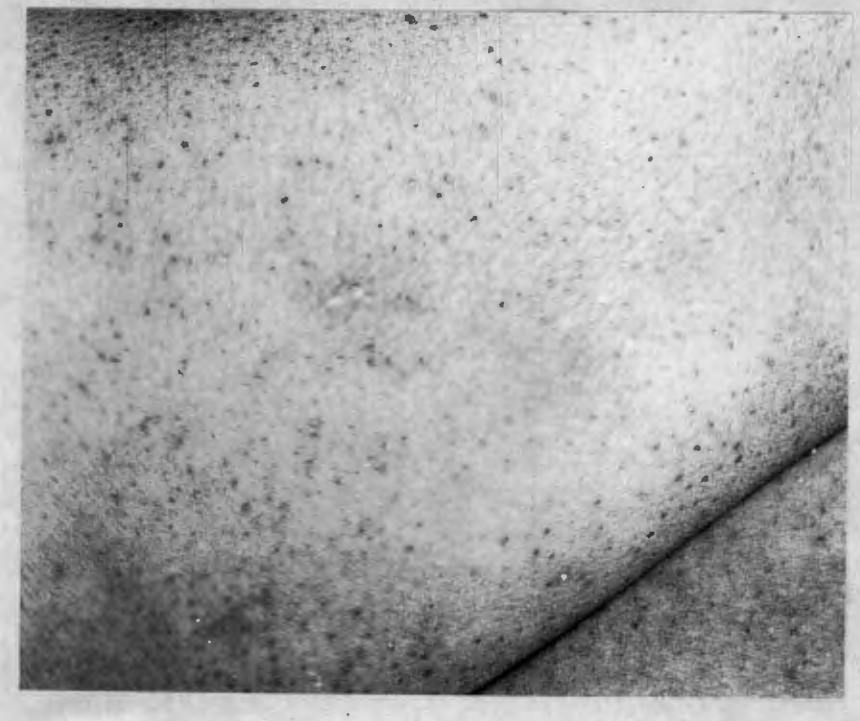


FIGURE 7

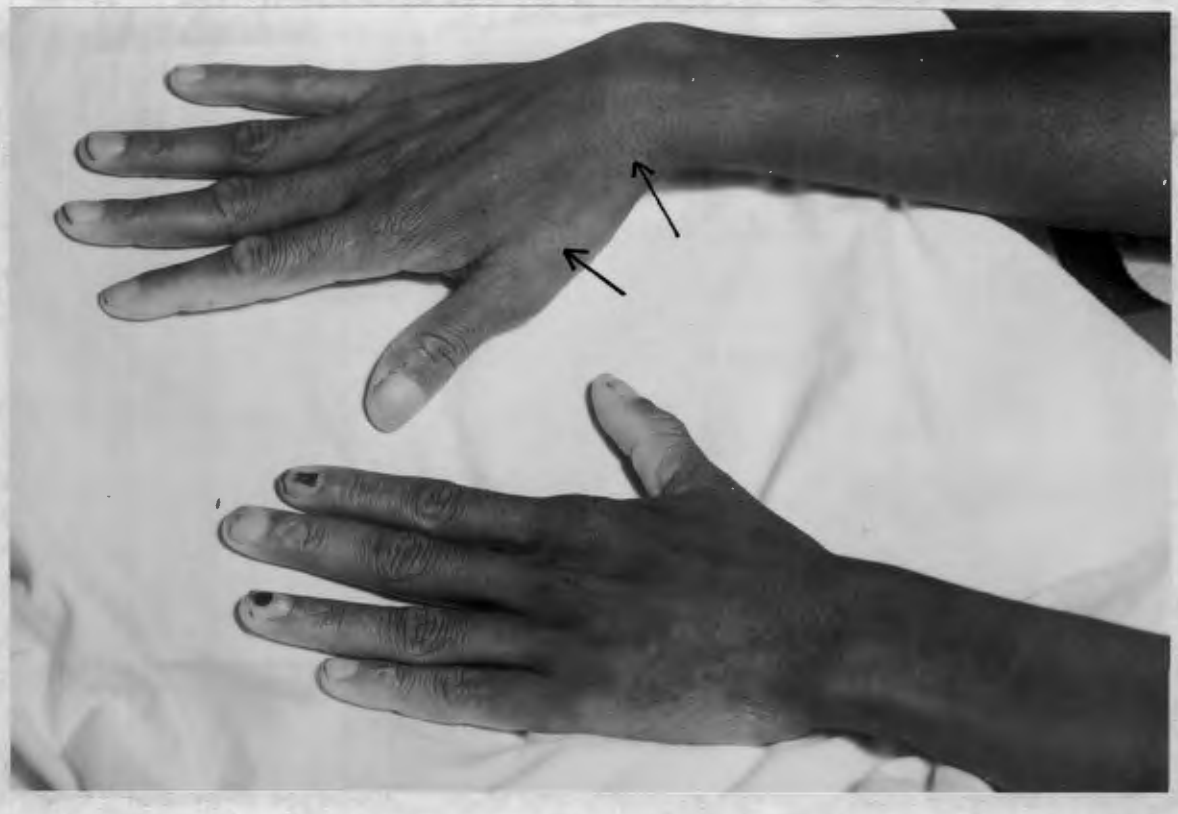


FIGURE 8

especially noticeable on the dorsum of the hand and forearm. This case (Case 6) also shows splinter haemorrhages of the nails. This pigmentation has been commented upon by PETERS et al (1948).

(c) Other skin lesions.

(i) CRANDON et al (1940) noted that at the time of appearance of the follicular hyperkeratosis the skin became dry and rough with the pores standing out in an exaggerated fashion, seen particularly on the extensor surface and backs of the hands. This has also been noted by RALLI and SHERRY (1941). Others [FOX et al (1940), PETERS et al (1948)] have denied its presence. This dryness was apparent in the cases of this series and many cases remarked on their improvement by the return of the normal "shine" to their skins. It may be this fact which has produced such adjectives as dirty, muddy, unwashed or dull to the complexion usually associated with scorbutics. [DRY (1933), RALLI and SHERRY (1941), VILTER et al (1946), JENNINGS and GLAZEBROOK (1938)]. "Unwashed" is a good description for the complexion of the cases in this series.

Other early skin changes mentioned [PETERS et al (1948)] occurring after the appearance of petechiae are:-

(ii) Recent scars became red and livid and returned to normal after about two months of treatment;

(iii) Exacerbation of acne present before the experiment began.

(iv) Later during the course of the human experimental work [PETERS et al (1948), CRANDON et al (1940)] the phenomenon of delayed wound healing was seen. That this is a feature of scurvy has been commented upon by various authors. [DRY (1933), RALLI and SHERRY (1941), McMILLAN (1943), McMILLAN and INGLIS (1944)]. In this experimental work, even three months of total deprivation of vitamin C gave perfect wound healing. In both experiments [CRANDON (1940), PETERS et al (1948)] it is stressed that this phenomenon is only encountered when, and not before, follicular hyperkeratosis has been present for some time (three weeks - PETERS et al (1948)). This feature was of interest to this series as one case (Case 1) was diagnosed as having an epitheliomatous ulcer just anterior to the left medial malleolus, as it was a progressive ulcer of one month's duration with everted edges. Biopsy however revealed chronic inflammatory changes. This fact associated with his severe anaemia led to his admission to a medical ward diagnosed as haemolytic anaemia. In his history however he remarked that a shoe had abraded his skin at that area and this large ulcer resulted. It healed rapidly and completely within three weeks of the commencement of ascorbic acid therapy.

2. Gum changes.

Gum changes usually present themselves much later than the skin manifestations yet they are regarded as one of the most characteristic early diagnostic criteria of scurvy. After five months of deprivation of vitamin C, Crandon's gums were pronounced as normal [CRANDON (1940)] by a competent dentist. Three weeks after the first sign (follicular hyperkeratosis) the gums were slightly more boggy than usual but no other gross change could be seen. On brushing the teeth there was no trace of bleeding at any time. However X-Rays of the teeth demonstrated occasional "discontinuities in the lamina dura". Immediately after the fatigue test while in the scorbutic state the subject did show for the first and only time a small haemorrhage at one gingival margin. Their conclusions, supported by the lack of changes in the edentulous, were that gum changes were not a true scorbutic sign but depended on - even in part - gingivitis, dental caries or both.
These facts, i.e. absence of changes in the edentulous and the state of dental hygiene influencing the time of their appearance and the rapidity of their progress, has been stressed by many [YOUNG (1938), SHAFAR (1949), VILTER et al (1946), NIELSON et al (1937), SHULTZER (1933)].

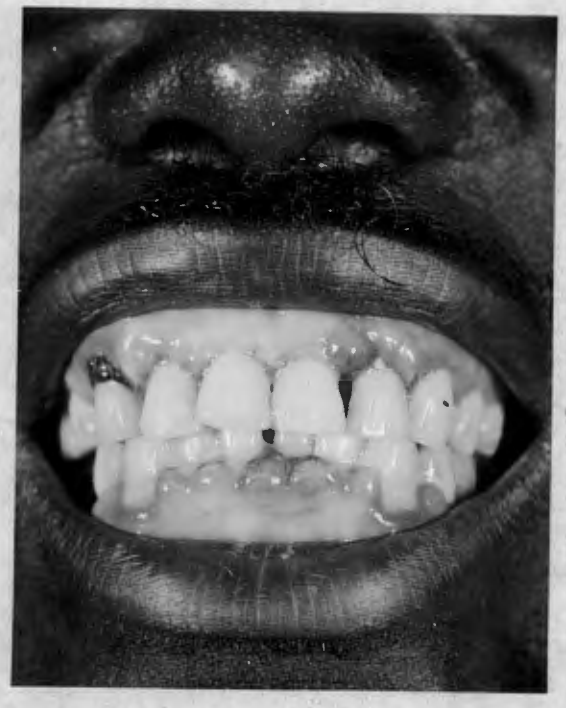
In the Medical Research Council experiment [PETERS et al (1948)] the earliest signs noted were tiny haemorrhages in and swelling of the tips of the interdental papillae. This was seen after twenty-six weeks of deprivation of vitamin C by which time all the volunteers had follicular hyperkeratosis.

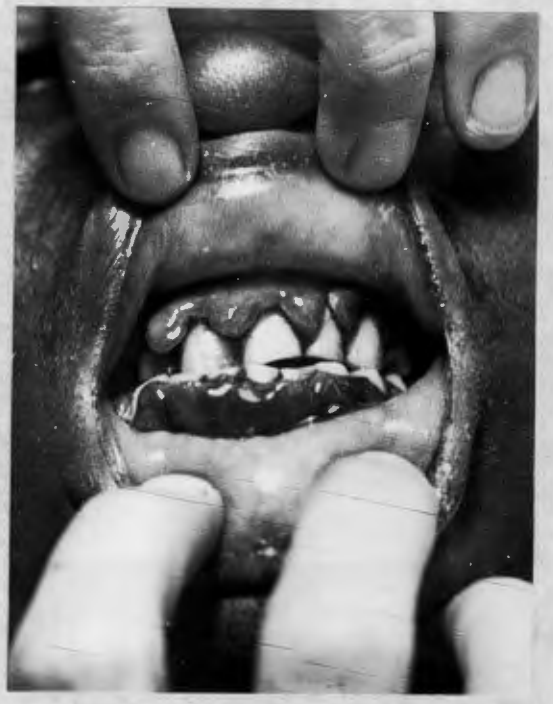
Nine of the ten volunteers had developed gum abnormalities within the following two months. The most severe changes were seen in two volunteers who had evidence of gingivitis and parodontal disease at the start of the deprivation. Ascorbic acid therapy for ten to fourteen weeks had to be given before resolution occurred. ANDREW (1949) developed one ulcer on the gums and another on the hard palate one month after the skin lesions appeared. No bleeding occurred. These gradually disappeared within two months of ascorbic acid therapy. GOTTLICH (1940) mentions that the actual condition of the gums in clinical scurvy was not the picture of profuse stomatitis but was rather localised to certain areas, the remainder of the gums often being apparently quite unaffected. This was noted in the case described by YOUNG (1938). The presence of haemorrhages in only those portions of the gums surrounding teeth is very characteristic [NINENSON et al (1937), SHULTZER (1933), VILTER et al (1946)] especially deformed or broken teeth [SHAFAR (1949)]. As the duration of scurvy increases the gums become more swollen and boggy and oozing of blood will follow even minor trauma [SHAFAR (1949)] but rarely are more than a few ml. of blood lost a day [VILTER et al (1946)]. The gingival enlargement may become so pronounced that it even covers or hides the teeth. Ulceration, infection and even gangrene [SHAFAR (1949)] may ensue resulting in a foul breath [VILTER et al (1946), JENNINGS et al (1938)]. Mastication becomes difficult and painful and the deficiency of vitamin C is aggravated further

by reduced food intake [VILTER et al (1946), SHAFAR (1949)]. Teeth become loose and may even fall out [RALLI et al (1941), SHAFAR (1949)]. For the frequency of gum changes one may again refer to the larger series. McMILLAN and INGLIS (1944) without giving figures state that where teeth existed, spongy and infected gums were very common. Eight had bleeding gums but this was present only in association with gross changes elsewhere. RALLI and SHERRY (1941) noted it in twenty-three of their twenty-seven cases. FOX and DANGERFIELD (1940) found changes in only twelve of their twenty-eight cases - in two cases these were the only features of scurvy, whilst VILTER et al (1946) found no changes in their edentulous patients, but in thirteen of the fourteen with teeth, all degrees of gum change were found. They stress the blue red colour of the affected gums even in the presence of a haemoglobin level of 6 or 7 G./100 ml. DRY (1933) mentions its frequency and comments on the frequent carious teeth found, which he regards as unusual as the African aborigines are credited with teeth as near perfect as can be found.

Even the Bantus of the older age group had all their teeth in this series and all degrees of gingival change were seen from slight interdental hypertrophy, to a degree where the lower teeth were nearly hidden, to large tender friable blue red excrescences on the palatal side of the alveolar margin (see Figs. 9 to 13). In all but one of the

thirty-two cases some degree of change was present. In two cases superficial examination of the teeth showed the gums to be normal but over two lower molars some interdental gingival hypertrophy was apparent. Pressure on the areas which appeared normal was tender and one of these two stated that bleeding occurred when he brushed his teeth.





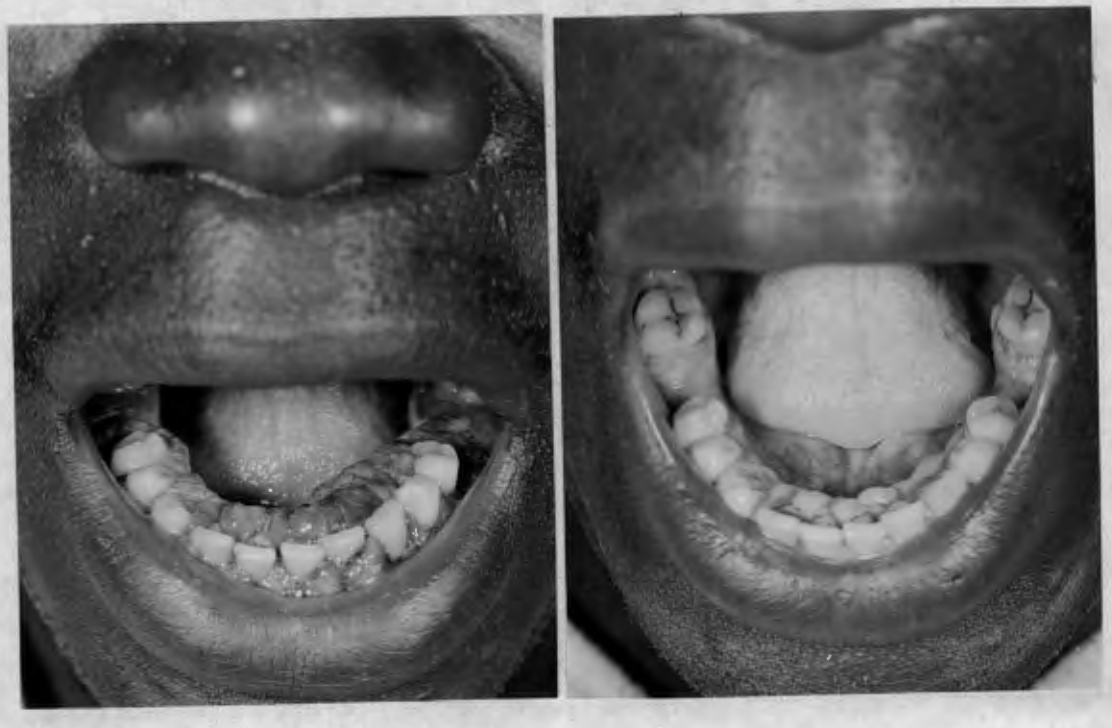
FIGURES 9,10,&11. To show the varying degrees of gum change in scurvy

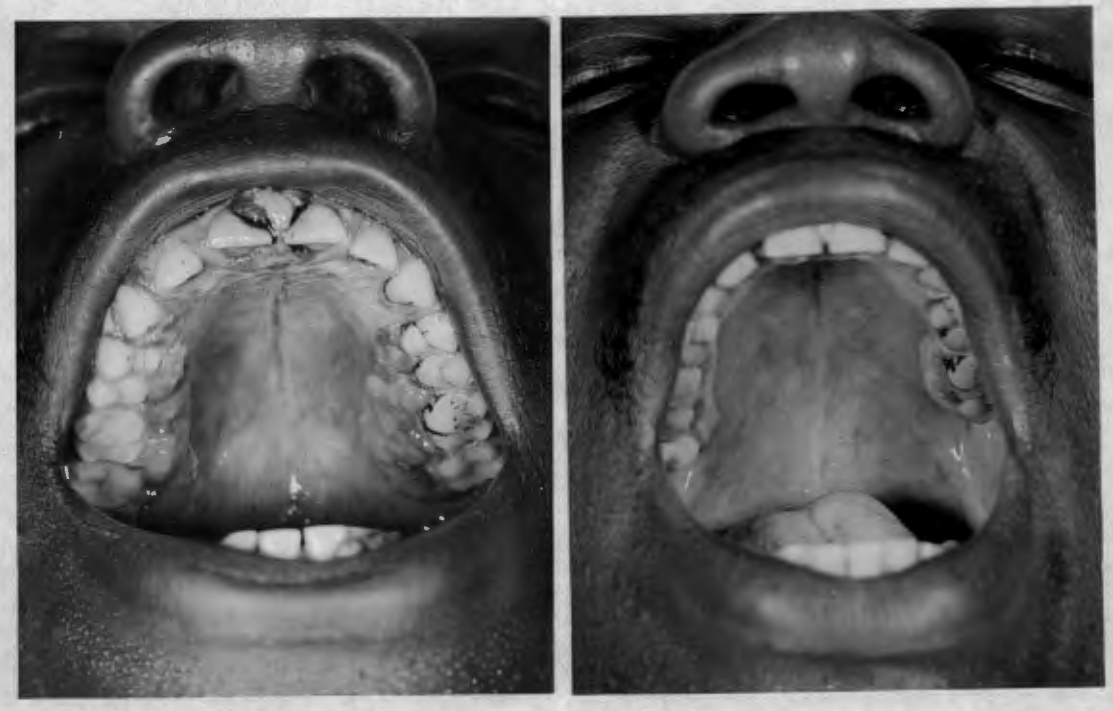
FIGURES 12 and 13

The effect of Ascorbic acid on gum lesions of scurvy

(a) before

(b) 12 days later





Twenty-five of the thirty-two cases complained of pain in the mouth. The only two who did not, in the "Personal" series, were those who had localised minimal gum changes.

Obviously carious and loose teeth however, were found only in nine of the thirty-two cases. In this series it appeared to be the larger and more friable lesions that responded the quickest to ascorbic acid therapy, whilst as noted before [PETERS et al (1948)] the final disappearance of the gum hypertrophy took more than two months treatment in some cases, but marked improvement was usually seen within ten days.

Diffuse gingival hypertrophy is by no means characteristic of scurvy. Pyorrhoea with associated gingival hypertrophy is frequently seen in the Bantu. Due to this swabs were taken of the gums in a few of the personal series. Attempts with antiseptic mouth washes and penicillin lozenges to clear up the infection were successful in that the foetor and pain was much improved. The gingival hypertrophy did decrease in size slightly but did not revert to normal until vitamin C was given,even after a long preliminary control period.

This fact, together with the fact that the absence of gum changes in the edentulous and that the gum change may be present only in localised areas with the other teeth and gum margins apparently normal, makes this sign less reliable for the confirmation of the diagnosis of scurvy. Its presence due to pyorrhoea or other causes adds to this. The value, then,

of follicular hyperkeratosis in deciding on a gum lesion of doubtful origin or of any other possible scorbutic sign, is obvious.

3. Deeper Haemorrhagic Manifestations.

Only in one case in experimental human scurvy did the experiment proceed long enough for the above to become manifest. After seven months he developed effusions into both knee joints and ecchymosis after a long walk [PETERS et al (1948)]. DALLDORF (1931) however shows the increasing tendency for haematoma formation to occur as the disease progresses. The specificity of these lesions in scurvy is shown by their response to pure ascorbic acid in both the guinea-pig and man. RALLI and SHERRY (1941) describe that with progression of the disease large haematomata form, leading to hard brawny tender swellings, termed "scurvy Sclerosis" by HESS (1920) with the skin over it becoming glossy and oedematous. As they become more extensive they are accompanied by oedema in the extremities. Eight of their twenty-seven cases had massive haematomata. Four of VILTER et al's series (1946) had deep subcutaneous or intramuscular haemorrhages giving rise to tender deep masses stretching the overlying skin. Seventeen of their nineteen cases had ecchymoses varying up to twenty cms. in diameter, most commonly found around the knees, ankles, wrists and popliteal spaces. Hard brawny and extremely tender haematomata are regarded as characteristic by NINENSON and COHEN (1937). McMILLAN and INGLIS (1944) noticed that of their fifty-three cases, fifteen had deeper haemorrhages in one

leg, twenty-three in both legs and thirteen in both arms and legs. In isolated case reports [JENNINGS et al (1938)] extensive bruising and ecchymosis is described. BARNS (1947) describes a case with ecchymoses extending from the buttock to the foot with the knee joint grossly swollen and hot. All three of SHULTZER's cases (1936) had haematomata. On a vitamin C free diet and bed rest only, these gradually subsided and recurred on allowing the patients up. Rapid remission followed the intravenous administration of vitamin C. Reference has already been made [FOX (1940)] to the fact that stress modifies the site and extent of the lesion, e.g. more common in the arms in woodcarders and blacksmiths. McMILLAN and INGLIS (1944) also noted that a patient, confined to bed for six weeks, developed haemorrhages on the buttocks only, the legs being unaffected.

In the Bantu, DRY (1933) mentions the extensive bleeding into the subcutaneous, subperiosteal and intermuscular spaces, leading to the severe crippling and pain associated with the disease. He goes on to say that a native with a limp during an epidemic is probably scorbutic. FOX and DANGERFIELD (1940) found haemorrhagic induration of the muscles and subcutaneous tissues in nineteen of their twenty-eight cases. The affected muscle was hard and rigid, allowing hobbling only and the skin over the area was dark and hard. When near a joint, an effusion into the joint was noted.

In this series large haematomata in one or other

leg, with tense tight glossy oedematous skin - often with "peau d'orange" appearance - overlying them, was seen in twenty-six cases of the total series of thirty-two. They were very tender and warm, and in three cases X-Rays were taken in the Out-patient department to exclude cellulitis from an underlying osteitis. The site was in most cases the popliteal fossa extending down into the calf giving the fleshy part of the tendo Achilles a "boggy" feel (see Fig. I4). Five of these twenty-six cases had haematomata in both legs, and three of the remainder had subperiosteal haemorrhages in addition, two on the tibia and one on the ulna. Only one attributed the cause to trauma. Of the twenty-six cases nine had additional haematomata in the arms or forearms. Nine of the twenty-six cases had pitting oedema of the ankle distal to the haematoma. In four cases an area of

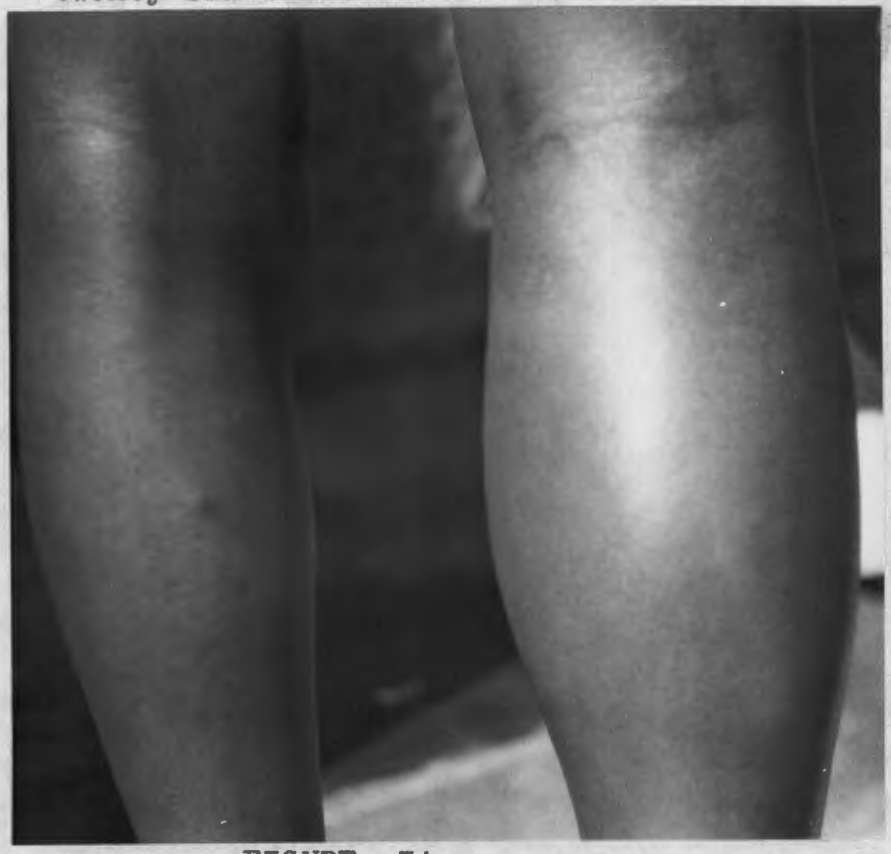


FIGURE I4

increased pigmentation of the skin was obvious over the haematoma. One of the above cases, the only Cape Coloured in the series, had, in addition, visible subcutaneous ecchymoses of the thigh and legs.

One case not having large intramuscular haematomata had, instead, diffuse ecchymosis throughout both thighs and legs. His black skin made recognition of the bruising difficult and the lines of demarkation of the ecchymoses became more obvious after twenty-four hours in bed. This was an unusual finding in the Bantus of this series and appeared to resemble, in palpation and appearance, McMILLAN's description of "Scurvy Pseudoscleroderma" (1943).

Five of the above mentioned twenty-five cases with haematomata had gross but non-tender swelling of the neighbouring knee joint. Two of these were aspirated and a clear, straw coloured fluid was withdrawn. VILTER et al (1946) report, however, the aspiration of blood from the knee joint of one of their cases. Consequently scurvy must be coupled with trauma and haemophilia as being a cause of frank haemorrhagic joint effusions.

RALLI and SHERRY (1941) and SHAFAR (1949) mention that as the patient is prone to infection, these haematomata are liable to suppurate. This is of interest with respect to the close similarity between scurvy and suppurative tropical myositis (GELFAND)*. No case in this series showed signs of abscess formation. Furthermore neither of the two cases of

*(1944)

tropical myositis seen here during the last year had any of the typical skin or gum changes associated with scurvy.

Splinter haemorrhages of the nails has been observed by VILTER et al (I946) and WOOD (I935). In this series splinter haemorrhages were noted in two cases. (see Fig. 8 page 68.)

Epistaxis, noted in one case in this series, has received frequent comment as a manifestation of scurvy (RALLI and SHERRY (I94I), SHAFAR (I949), EDDY and DALLDORF (I94I), McMILLAN and INGLIS (I944).

Haemoptysis together with haematemesis (HATHERLY (I947)), haematuria (RALLI and SHERRY (I94I), SHAFAR (I949)) and blood in the gut and bronchi at post-mortem (GORDON (I942)) have all been reported. Melaena with intestinal colic has been observed (McMILLAN AND INGLIS (I944)) and haemorrhages into serous cavities, such as the pericardial and pleural spaces (SHAFAR (I949)) or the meninges of the brain (GORDON (I942)) may occur. No case in this series had visibly detectable haemorrhages in or from these areas. Retinal haemorrhages were found in three cases of this series but these may have been due to the associated severe anaemia.

This haemorrhagic tendency, particularly into the deeper tissues, is probably the cause of the pain experienced by scorbutics. McMILLAN and INGLIS (I944) stress pain as the first symptom that brings the patient to the doctor. Lumbago,

which they attribute to haemorrhages into the back, came on before pain in the legs in three of their cases. Discomfort was usually mild at first but became progressively more severe. It appeared first in one leg, then the other and then the arms. Within a month most patients were crippled and were pleased to be allowed to rest in bed.

VILTER et al (1946) state that their patients dated the onset of their illness to one or two months before. Dull aching in the legs was usually the first symptom, followed by swelling, pain or discoloration around the knees or ankles. Only later did those with natural teeth complain of painful swollen gums, which interfered with mastication or from which blood oozed.

Of the total number of thirty-two cases in this series pain, with or without stiffness in one or both legs, was complained of in twenty-seven. In the personal series, nine of the eleven who had pain in the legs, had haematomata to account for this, whereas pain in the upper limbs, experienced by ten of the thirty-two cases, was present concurrently with haematomata in eight. Diffuse sheets of haemorrhage along the fascial planes may have been present, however.

4. Pallor.

The early writers on scurvy noticed how frequently patients with scurvy are pale and anaemic looking [MACVICAR (1906)]. Many in fact considered this as due to some circulatory change present, and not due to anaemia. On the

other hand VILTER et al (1946) have remarked that the gums may be blueish red even with a haemoglobin level of 5 or 6 G. per 100 ml. That anaemia is a frequent occurrence in long standing scurvy is now well recognised [VILTER et al (1946), CARTWRIGHT (1947), SHAFAR (1949), METTIER et al (1930), JENNINGS et al (1938)]. As this will be discussed fully later, only brief mention will be made here, that of the total number of thirty-two cases, pallor was noticed as a feature in eighteen.

5. Cardio-Vascular Effects.

(a) The Blood Pressure.

In Crandon's experiment [CRANDON et al (1940) his blood pressure was 120/70 mms. Hg. until after the fourth episode of blood loss of about 500 ml. This venesection took place after the development of the first clinical sign of scurvy. The blood pressure then fell to 90/60 mms. Hg. During the next week the systolic pressure rose to 98 but never exceeded this level until ascorbic acid was started, when it promptly rose. At this time he was still on the same vitamin C-free diet, consequently they consider this as significant. The blood volume was normal but there was a fall in blood sodium at the end of the experiment, which rose after ascorbic acid therapy. Pertinent in this respect, they state, are the facts that scorbutic guinea-pigs have diminished adreno-cortical hormones and at autopsy adrenal cortical atrophy. Pertinent here too, then, is the mention of

Table 3.

Case No.	Age (Yrs)	P.C.V. %	Blood Pressure (mms. Hg.)	
			Systolic	Diastolic
1.	30	14	110	55
2.	37	37	130	90
3.	40	20	105	50
4.	49	23	135	85
5.	25	22	126	68
6.	28	15	130	70
7.	50	53	180	140
8.	35	22	105	60
9.	24	23	130	75
10.	60	26	110	70
11.	27	42	110	70
12.	55	27	125	70
13.	59	15	140	80
14.	60	51	110	70
15.	32	42	110	70
16.	37	45	170	110
17.	49	16	135	70
18.	50	31	125	70
19.	30	39	150	85
20.	35	26	105	50
21.	30	14	110	60
22.	25	17	120	80
23.	35	24	120	80
24.	52	25	100	60
25.	25	8	105	60
26.	53	12	110	55
27.	28	1.8*	110	60
28.	30	28	130	65
29.	45	14	120	70
30.	45	2.8*	115	85
31.	36	4.3*	140	84
32.	22	4.0*	115	70

* Red Blood Cells in Millions per cubic m.m.

acute vasomotor collapse that may occur in scurvy giving a syndrome akin to that described, in haemorrhage into the adrenals [VILTER et al (1946), SHAFAR (1949)]. In Vilter et al's series (1946) the blood pressures, frequently low on admission, fell further in some cases on a diet free of vitamin C and low in vitamin B complex. Two fell to very low levels - one as low as 60/20 mms. Hg. followed by death in the one (aged 75 years) and a cerebral thrombosis in the other (aged 54 years). Both these patients were severely anaemic. All these cases showed a progressive return to normal of the blood pressure with the administration of vitamin C.

Unfortunately most of the other larger series and isolated case reports are on observations in elderly men. Consequently one is not surprised to hear arteriosclerosis cited as a possible predisposing cause [METTIER et al (1930)]. Janet Vaughan's case (1934) aged 71 years had a blood pressure of 120/60 mm. Hg., and X-Ray examination showed excessive calcification of the arteries.

In Table 3 the blood pressures of the cases in this series taken on admission have been compared with the patient's age and packed cell volume. Eleven of these patients had diastolic pressures below 70 mm. mercury and all eleven were grossly anaemic. Two patients had hypertension but both had normal packed cell volumes.

Unfortunately at this time CRANDON et al's (1940) work on sodium levels was not available so no adequate follow-up, or sodium determinations were performed to decide whether the lower blood pressures were a scorbutic feature or merely due to the associated anaemia in these cases.

(b) Cardio-Respiratory Symptoms.

Dyspnoea on exertion and dizziness have been mentioned by investigators in their series [RALLI et al (1941), VILTER et al (1946)] and in isolated case reports [JENNINGS et al (1938)]. How much of this is cardiac in origin or due to the associated anaemia is hard to assess. Most of the above cases are in the arteriosclerotic heart disease age whereas the majority had showed anaemia of some degree.

In the total series presented here, four cases were admitted as collapse cases - their packed cell volumes were 8%, 12%, 15% and 15%. One had been confined to bed for a fortnight and noticed that whenever he tried to assume the erect position he lost consciousness. Three patients complained of dizziness and dyspnoea on exertion - all anaemic. One of these - the collapse case with 12% packed cell volume - also remarked on palpitations and nocturnal dyspnoea. His age was 53 years. The other two were the oldest patients in the series aged 59 and 60 years. One had electrocardiographic evidence of previous myocardial damage

but also had a packed cell volume of 15%.

(c) Cardiac Enlargement.

SHAFAR (1949) mentions that this may accompany the scorbutic state and that haemorrhage may occur into the myocardium and pericardium. FOX and DANGERFIELD (1940) report, in their series of twenty-eight cases, one case of congestive cardiac failure. One of GOTTLIEB's (1945) four cases died five days after admission in congestive cardiac failure. He was grossly anaemic, however. SHATTUCK (1928) also reports a death from congestive cardiac failure in an elderly patient. METTIER et al (1930)state that cardiovascular damage, chiefly arteriosclerotic in type was present to a greater or lesser degree in all of their eight cases of scurvy in adult males. As mentioned previously, most of the case reports in the literature are on males over 50 years of age. The series presented here were mainly in the younger age group (20 to 40 years) and no case showed cardiac enlargement detectable clinically or on X-Ray of the chest. One case, (Case 7) aged 50 years, however had a small aneurysm of the ascending aorta shown on the X-Ray of the chest. His Wassermann and Kahn reactions were negative but his Berger was doubtful.

(d) Abnormality of the heart sounds.

Soft blowing systolic murmurs were heard in fifteen of

the thirty-two cases. All these cases had gross anaemia and the murmurs were not heard when the anaemia had been corrected.

(e) Electrocardiographic changes.

Much anxiety was aroused by the condition of one of the ten human volunteers who developed scurvy [PETERS et al (1948), British Medical Journal (1948)]. Nineteen hours after heavy physical exercise he had severe pain in the lower sternum, with dyspnoea, cyanosis and drop in blood pressure. The electrocardiogram showed high ST levels in leads I and II. The pain passed off after nine hours. Another subject developed praecordial pain and an electrocardiogram demonstrated a partial heart block with a PR interval of 0.28 second. However long after treatment the PR interval still varied between 0.20 and 0.28 second depending on posture, breathing, administration of drugs. The authors did not feel, therefore, that these changes had any connection with the deficient diet. However, it is advisable [British Medical Journal (1948)] for any patient suffering from scurvy to be admitted to hospital as soon as possible.

Nowhere else in the literature on scurvy has there been mention of coronary thrombosis occurring even though most case reports are in elderly arteriosclerotic males. If one accepts the pathogenesis as thrombosis occurring

on an atheromatous plaque under which a haemorrhage has occurred during a period of exertion, then surely with the increased bleeding tendency found in scurvy coronary thrombosis should be more commonly found. However as scurvy progresses the disease itself by the formation of haematomata offsets any attempt at undue physical exertion, and it is only in the milder cases that such may occur, i.e. when they are subjected to fatigue tests. Nevertheless the Bantu here continues his heavy physical labour long after haematomata have formed and apparently has no ill-effects. The Government Pathologist who has to perform autopsies on unexplained sudden deaths will testify as to the rarity of a case of scurvy being found. Consequently one wonders whether the case mentioned above with ST elevation in leads I and II had anything to do with his mild scorbutic state.

VILTER et al (1946) record two electrocardiographs. One showed ST_1 and ST_4 depression and the other showed diphasic T, and inverted T_4. Both cases were over 65 years. In both cases reported by JENNINGS and GLAZEBROOK (1938) the electrocardiographs were normal.

Seven electrocardiographs were recorded in this series. All except two were normal. In one an inverted T in lead aVF became upright, without any rotational or axis change in position, after vitamin C therapy. He was

25 years of age. The other, aged 60 years, had inverted T waves in leads I, II, aVL, V_4, V_5 and V_6 with no hypertension or detectable left ventricular enlargement to account for these changes. The changes were assumed to be due to an old antero-lateral myocardial infarction [ORAM (1949)]. Very soon after admission he complained of sudden pain in the chest. His blood pressure dropped from 140/80 to 110/70 mms. Hg.; he became unconscious and despite heroic measures, died. Unfortunately no post-mortem was obtained but in view of the previous electrocardiographic change and the clinical features of the attack, coronary thrombosis was accepted as the mode of death.

The doubt that a relation exists between scurvy and coronary thrombosis has been expressed already. For record purposes however, there is now suggestive evidence that features suggesting coronary thrombosis occurred, as it were, in two extremes of scurvy - in one a mild scorbutic after severe physical strain [PETERS et al (1948)] and in the other a severe scorbutic resting in bed, with severe anaemia.

(f) Oedema of the ankles.

Bilateral oedema of the ankles has been reported in scurvy. SHATTUCK (1928) attributed this as due to the accompanying anaemia or cardio-vascular damage. The latter was suggested, as frequently scurvy develops in the elderly

age group. Bilateral oedema of the ankles was noticed in six patients of the total series. Unilateral oedema distal to the haematoma was very common, on admission. In the six cases mentioned above, bilateral haematomata were present in five. The sixth had very severe anaemia.

(g) <u>The diuretic effect of Ascorbic Acid.</u>

ABT and FARMER (1938) mention this as a pharmacological action of ascorbic acid. ABBASSY (1937) caused a slight diuresis in normal non-oedematous patients whilst EVANS (1938) induced a greater diuresis with ascorbic acid than with digitalis in oedematous cardiac patients. This diuretic effect was less than that of theobromine or ammonium chloride, however. SHAFFER (1944) found a greater diuretic effect when this vitamin was given orally to patients with heart failure, than when given intravenously. Furthermore he felt that ascorbic acid enhanced the efficacy of a mercurial diuretic.

Mention has been made of the experimental work in guinea-pigs whereby symptoms could be retarded and minimized by withholding water. Furthermore a drop in sodium levels, a normal blood volume and a drop in blood pressure in human scurvy has just been discussed.

In this series a decreasing urinary output was noted as the disease progressed despite a more or less constant intake

FLUID BALANCE GRAPHS

Represent the difference between the urinary output and fluid intake (in Ozs) - Diet constant; no excess sweating; no diarrhoea.

FIGURE 15

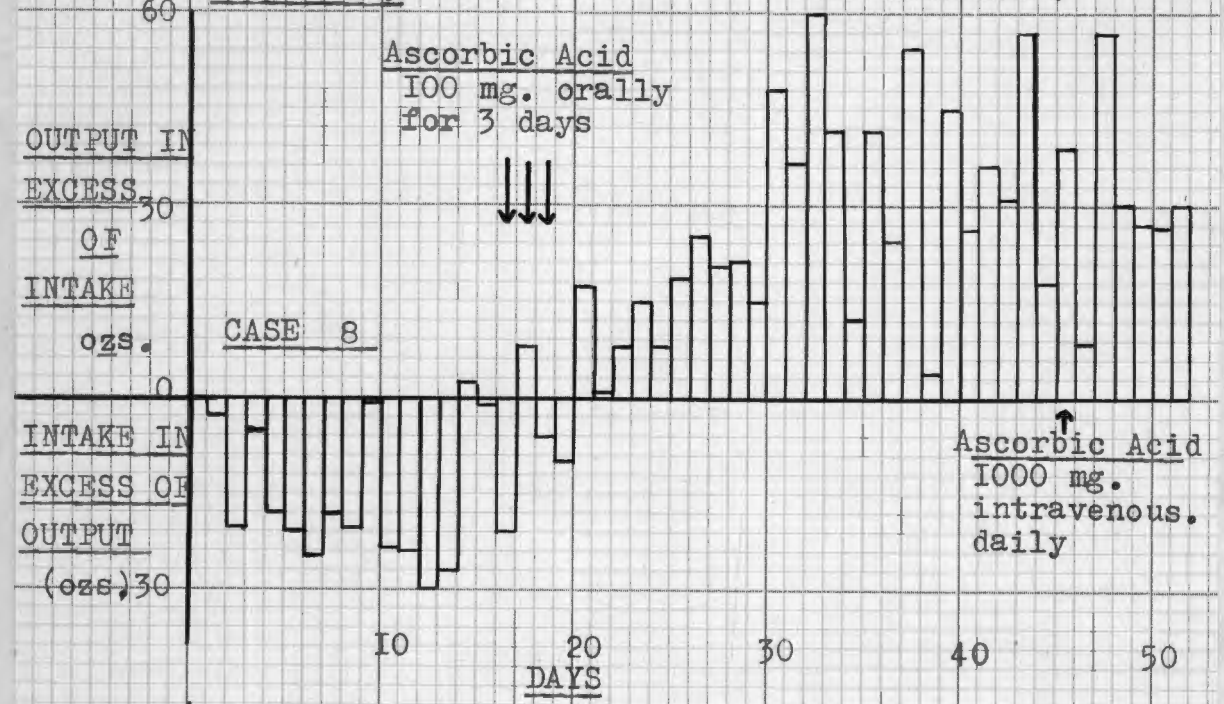
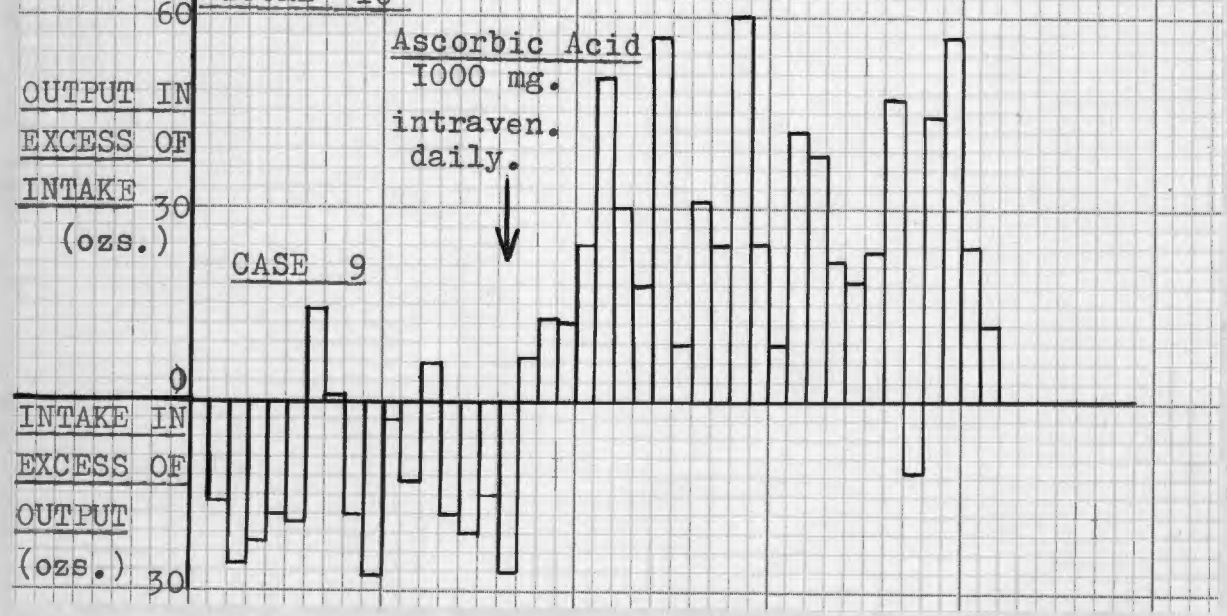


FIGURE 16



and bed rest.

The first case observed from this aspect dropped as low as seven ounces in a day. Increasing the intake, which did not please the patient, led to only a slight rise in output. After ascorbic acid was given (intravenously) the output never fell below forty ounces a day. In two other cases an insuffieient period of control elapsed before treatment to exclude bed rest alone as the cause of the diuresis. In one of these, the urinary excretion was 28 ozs., 17 ozs., 17 ozs. and 28 ozs. for the four successive days prior to treatment. On the fifth day after intravenous ascorbic acid therapy (1000 mgm. daily) and for the succeeding twelve days the output never fell below 50 ozs. and was frequently above 70 ozs. daily despite a more or less constant intake of 35 to 40 ozs.

In three more patients the fluid balance was studied in detail. Figures 15 and 16 illustrate graphically this water retention and diuresis after ascorbic acid. In the third case, a mild scorbutic with no anaemia, no such significant diuretic effect was noted. Figure 15 shows that 100 mg. ascorbic acid, given orally for only three days was sufficient to cause this increased excretion and one month later when 1000 mgm. intravenously was given, no further response was noted. Figure 16 shows that intravenous ascorbic acid (1000 mgm.) produces a response as good as the oral route and somewhat sooner,

but different doses were used, so no accurate comparisons between figures 15 and 16 can be drawn with regard to SHAFFER's (1944) views on the effect on different routes of administration.

Unfortunately this aspect of scurvy has apparently not been studied. Earlier on in the predisposing causes mention was made of possible diminished glomerular filtration as a means of conserving vitamin C in the body. It is also of interest to note that the pituitary and adrenal cortex have the highest concentration of ascorbic acid of all the organs in the body. As this aspect did not fall within the purpose of this thesis blood volumes and renal blood flow studies were not performed. The blood urea recorded at this time in these cases was always within normal limits and no albuminuria occurred. No case was in cardiac failure. Further elucidation of this interesting aspect would be difficult as each case in which this water retention was noted had severe anaemia. The mild scorbutic who showed no such effect was not anaemic. All cases however had some evidence of liver dysfunction by the chemical tests. It would be difficult, then, to regard this as a specific feature of severe scurvy despite the immediate response to ascorbic acid. Nevertheless as an interesting observation it was recorded.

6. Loss of Weight.

Despite this apparent water retention loss of weight occurs in scurvy. With the type of diet eaten by patients in this series it is not surprising.

It is well known that guinea-pigs made scorbutic, lose weight. Likewise CRANDON et al (1940) record a gradual and continuous loss of weight up to 27 lbs. in man. This was attributed to the type of diet as no gain in weight was noticed when only vitamin C was added to the same diet.

That this was not an inevitable accompaniment of the early stages of scurvy was shown by PETERS et al (1948). None of the ten volunteers lost weight.

In the personal series all patients had gained weight by the end of their hospital stay except two. Both these cases lost three pounds. Analysis of seven of these patients who gained weight during their hospital stay as measured by weekly weighings revealed the following facts:-

(a) Four gained weight, despite a diuresis, when vitamin C alone was added to the diet. All four gained further, but no more rapidly, when full hospital diet was given. The average gain per week was almost three pounds.

(b) The other three cases registered approximately the same weight on discharge as on admission. One lost four pounds during the time he was getting

the basic diet plus vitamin C and gained five pounds on the hospital diet. Another did not gain until he had full hospital diet and the third lost the weight he had gained after a month of hospital diet.

Some evidence then points to weight loss occurring in scurvy per se, when the diuresis is taken into account. Accurate conclusions cannot be drawn however.

7. Pyrexia.

The pyrexia, that may occur in scurvy, was a cause for comment by earlier writers [MACVICAR (1906)] and later [EDDY and DALLDORF (1941), VILTER et al (1946) and SHAFAR (1949)]. It has never been adequately explained. Pyrexia was not found in every case in this series. It was not present in those cases who were not anaemic. In those where it was present it varied between 99°F. and 101°F. In those studied over long periods of control prior to treatment, the pyrexia remained and would disappear within five days of vitamin C therapy. It did not appear to be related to the extent or even presence of haematoma formation. With rest in bed these slowly absorbed but the pyrexia persisted. Suppuration did not occur in this series and the strict relation to vitamin C therapy made undetected underlying infection unlikely. It was felt that the accompanying anaemia, with its immediate response to ascorbic acid, could explain the pyrexia.

Evidence of other Deficiencies or Diseases Co-existent.

1. Evidence of co-existent infections or diseases.

The question of the pyrexia in scurvy as being due to co-existent infections has been mentioned. The presence of dental caries in association with the gum changes has received comment. In Chapter II mention was made that infection may shorten the incubation period of scurvy or speed its progress. Finally, some have reported that the scorbutic is less resistant to bacterial or viral invasion.

(a) Splenic enlargement.

As mentioned under predisposing causes, of the total series of thirty-two cases, nine, which included three of the personal series of thirteen cases, had a splenomegaly palpable one to three cms. below the left costal margin of very firm consistency. All these cases were East African Bantus where malaria is endemic. In no case could malarial parasites be demonstrated in the peripheral blood or bone marrow and no case gave any story compatible with active malarial infestation. The size and consistency of the spleen did not change in any case, after full recovery. Consequently it was felt that the diagnosis was probably the splenomegaly of chronic inactive malaria and played no part in the present illness.

(b) Tuberculosis.

Under predisposing causes mention is made how routine chest X-Ray examinations were made on each of the personal series of thirteen cases. One case had right apical pleural thickening with an infraclavicular lesion suggestive of inactivity. Several sputum examinations failed to reveal acid-fast organisms.

In the total series of thirty-two cases, one case with a normal chest X-Ray and negative sputa examination had tuberculous adenitis of the left groin proved on histological examination. His sedimentation rate was normal on discharge six weeks after admission. Another case had bilateral hydrocoeles of five weeks duration. On tapping a clear straw-coloured fluid was obtained. The underlying epididymi were enlarged, non-tender and "craggy". Pyelography and X-Ray chest was normal and twenty-four hour urine specimens contained no acid-fast organisms.

(c) Syphilis and other Venereal Diseases.

In twenty-six patients the serological test for syphilis was negative, one was positive and no records are available for the test in the remaining five patients. The one case with the positive serological test was the patient with the tuberculous gland mentioned previously. One patient with two negative serological

tests (Wassermann and Kahn) aged 50 years, with a blood pressure of 180/140 mms. Hg. had early aneurysmal dilatation of the proximal aorta. His Berger test was doubtful.

One patient had a chancroid on admission.

(d) Evidence of Helminthiasis or other Intestinal Diseases.

Bacterial examination of the stools was performed routinely on each of the personal series of thirteen cases on admission. Tape worm (T.saginata) was found in one case. Treatment of this had no effect either way on the scorbutic features. Another case had intermittent attacks of diarrhoea with fresh blood in his stool. Bacteriological examination isolated no entamoebae or organisms. The condition was very mild, the patient had not noticed it, and it failed to respond to a course of sulphonamide therapy. No blood was seen again, once vitamin C therapy started. Possibly it was a scorbutic manifestation.

In the remaining nineteen cases of the total series, four stool examinations were performed and all were negative.

.2. Evidence of Liver Dysfunction.

Few reports exist on liver function studies in scurvy. WOOD (1935) reports a scorbutic female with an enlarged liver. WRIGHT and LILIENFELD (1936) devote a large part of their article-on the physiological and pharmacological effects of vitamin C-to the work at that time by German authors, on the relation between the serum proteins and vitamin C. These authors all reported an increase in albumen and total proteins in response to vitamin C therapy even in non-scorbutics. Wright could not confirm these results. Mention was also made that the sedimentation rate decreased. The part played in this mechanism by the liver was not apparently assessed, however.

CRANDON et al (1940) noticed a drop in total proteins as the disease progressed and a return to normal very soon after therapy. Unfortunately no differential protein estimations or liver functions are to hand to determine whether liver dysfunction or dietary lack was the cause of this hypoproteinaemia.

SHAFAR (1949) mentions fatty livers in experimental scurvy of guinea-pigs but the part played by any associated anaemia is not discussed.

VILTER et al (1945)(1946) found high urinary urobilinogen levels in their cases and felt that haemolysis was the factor, as those liver function tests performed

(Prothrombin times, Cephalin Cholesterol Flocculation and Bromsulfalein retention), were normal in most cases. High faecal urobilinogen levels supported their view. The extent of the ecchymosis, in their opinion, could not account for this. Unfortunately icteric indices and qualitative van den Bergh tests were used to assess the "jaundice" that was constantly present. In the discussion after the preliminary presentation (1945) C.J. Watson felt that hepatic dysfunction was more probable. Low serum albumen levels were also found but Vilter stated that they could be easily explained on the dietary protein deficiency.

The frequent finding of urobilinuria by the spectroscopic test in the cases of this series led to daily quantitative urobilinogen studies. Concurrently repeated serum bilirubin, serum protein estimations with the thymol turbidity and flocculation and serum colloidal gold reactions to assess liver function, were performed. The effect of bed rest alone while on the diet on which the disease developed was observed. Likewise the effect produced by an increase in the haematoma present,—by increasing the capillary pressure—and the effect of pure ascorbic acid only being added to this basic diet, was observed. This also allowed an assessment of the part played by the anaemia present.

A tabulated summary (see Table 4) expresses these results. The second column denotes the day before or after ascorbic acid therapy of the test. Before is designated

TABLE 4

EVIDENCE OF LIVER DYSFUNCTION

CASE NO.	DAYS BEFORE(-) OR AFTER(+) VITAMIN C	P.C.V. %	BILIRUBINAEMIA mg.%	SERUM ALBUMEN G%	SERUM GLOBULIN G%	ALB./GLOB. RATIO	THYMOL TURBIDITY	COLLOIDAL GOLD	THYMOL FLOCCULATION	SERUM CHOLESTEROL mg/100ml	URINARY UROBILIN(OGEN) mg/day	FAECAL UROBILINOGEN mg/100G	PRESENCE OF HAEMATOMA
1	+1	14	1				1	0	0		-		-
2	-1	37	3.2	4.1	3.9	1.05	4	4	3	81	++		-
	+40	40	Ict.Index 4							140	-		-
3	-1	20	Sl. trace	3.5	2.8	1.25	1.5	1	1		+		+
	+40	42	0	4.9	2.1	2.29	3	2	1		-		-
4	0	23	0	3.8	2.2	1.73	1.5	1	0		+		++
5	-6	22	0	3.3	2.6	1.27	4.5	5	4		+		-
	+28	40	0	4.5	2.5	1.80	4.5	5	4		-		-
	+120	46	0	4.5	3.0	1.50	4	2	1		-		-
6	-27	15	0	4.0	3.0	1.33	1	0	0		+		++
	-20	17	0	5.1	3.0	1.70	1	0	0		18.0		+
	-12	14	0	4.5	3.2	1.40	1	0	0		12.0	142.3	±
	-5	13	Ict.Index 28								57.0	402	++
	+5	20									5.0	205	+ ±
	+15	31	0	3.8	2.9	1.31	2.5	0	0		3.0	70.4	±
	+37	43	0	4.6	2.7	1.70	1	0	0				-
	+57	43	0				1.5	0	1				-
7	-25	58	0.5	5.3	3.2	1.65	4.5	4	4		19.6	77	-
	+21	53	0	4.6	2.7	1.70	7	3	3		6.5	55	-
8	-25	23	Sl. trace Ict.Index 14	3.4	2.5	1.36	2.5	1	1		59.9	427	++
	-22	20	0.5	3.6	2.0	1.80	2.5	3	1		21.7	480	++
	-7	22									3.7	210	±
	-1	22	0.6 (Ict.Index 22)	4.1	1.5	2.73	2.5	3	0	92	5.7	1140	+++
	+5	27	0	3.4	2.1	1.62	2.5	2	0		4.38	142.5	+ ±
	+17	35	0	4.2	2.1	2.0	4	3	1				±
	+39	40	0	3.5	2.7	1.29	3	4	3				-
	+42	41	0	4.8	2.1	2.29	4.5	2	2				-
9	-14	23	0				3.5	2	1		21.2	260	++
	-9	22	0 (Ict.Index 21)	4.6	2.4	1.91	4	2	1	109	6.85	182	+
	+5	26		4.7	2.6	1.81	4.5	3	3		4.2	92	±
	+12	29	0				4	3	2	78			±
	+36	40	0	4.0	3.4	1.18	4.5	3	3				-
	+39	41		4.6	2.9	1.60	3.5	2	1				-
10	-5	26	0	4.5	2.8	1.60	2.5	1	0		15.8	303	++
	+18	41					5.5	4	2		1.2		±
11	-7	42	1	2.8	3.7	0.76	1.5	0	0		20.8	395	++
	+7	45	0	3.8	4.2	0.90	3.5	0	0		6.7	300	+
12	-1	27	0	3.7	3.8	0.97	2.5	1	0		± 13.5	163	++
	+26	46	0	4.3	3.3	1.30	3.5	1	0		3.2		±
13	-4	15	0	4.5	3.3	1.36	2.5	2	1		63	510	++
19	+1	39	trace	4.7	1.4	3.36	1.5	2	0		+		++
20	+1	26	trace	3.6	3.0	1.20	1	2	0	101	+		++
22	+2	17		2.7	3.6	0.75	1	1	1		tr		++
23	+1	24	1.9	3.1	2.8	1.11					+		++
24	+2	25	0	2.8	2.3	1.22	2.5	1	0				+ ±
26	+1	12	1.2	2.6	2.4	1.08					+		+++

with the sign minus and after, plus. The last column denotes the extent and degree of haematomata present, designated by plusses. An attempt has been made with these signs to show the gradual absorption with bed rest alone whilst still on the diet on which the disease developed. The signs also show when an increase in the haematoma formation occurred. Full mixed hospital diet was only given when the packed cell volume was in the region of 40%. This statement only applies to Cases 2 to 12. Blank spaces in the table mean that the particular test was not performed. Plus or minus signs in column 11 (urinary urobilinogen) denote the results obtained with the spectroscopic test.

It will be seen than that in no case (except Case 1 who was not as intensively investigated), was there normal liver function as assessed by these chemical tests. Five of these cases had a hepatomegaly varying from two to five cms. below the ninth right costal cartilage, not hard, smooth and enlarged obliquely up across the abdomen. The features appeared to reflect fatty infiltration.

The cause of this liver dysfunction is not easily seen. Three possibilities exist:-

(a) A specific feature of vitamin C deficiency. The dysfunction should revert to normal with ascorbic acid only added to the diet.

(b) Dysfunction may be due, merely, to anoxaemia from

the associated anaemia. It, then, should be present only in those with the associated anaemia and should respond with the response in the anaemia.

(c) The lack of essential amino acids in the diet on which the disease developed.

Liver dysfunction due to a deficiency of vitamin C in the body acting by interfering with the utilisation of amino acids is a strong theoretical possibility.

SEALOCK et al (1941) noticed the excretion of tyrosine and its two principal deaminated products, parahydroxyphenylpyruvic acid and parahydroxyphenyllactic acid in scorbutic guinea-pigs when fed L-tyrosine. Proof that this metabolic disorder is primarily dependent on a state of ascorbic acid depletion, is afforded by the observation that the "tyrosyluria" is abolished when the vitamin is given orally or parentally.

Several years later [SEALOCK and LEPON (1948) and WOODRUFF et al (1948)(1949)] in confirming this noticed that liver extracts and especially pteroylglutamic acid (PGA) could modify this induced "tyrosyluria".

This work was later confirmed in infantile scurvy [MORRIS et al (1950)] and in adult scurvy [ROGERS and GARDNER (1949)]. The last mentioned authors studied four normals

and four scorbutics. Large amounts of "tyrosyl" derivatives appeared in the scorbutics' urines and vitamin C administration led to complete correction of the metabolic abnormality in 24 to 48 hours.

MORRIS et al (1950) feel that these observations do not imply a more complete utilisation of tyrosine and phenylalanine. PGA and ascorbic acid may produce these effects by rupturing the aromatic nucleus and thus non-phenolic residues would appear in the urine. They feel that other pathways of tyrosine metabolism, not necessarily dependent on ascorbic acid function, are in operation in vitamin C depleted infants.

They support this by noting these infants were capable of ingesting a formula which supplied 0.3 G. of aromatic amino acid per kilo of body weight without excreting abnormal quantities or urinary hydroxyphenyl compounds, even in the absence of ascorbic acid administration. Secondly, although phenylalanine is an essential amino acid these depleted infants showed no marked stunting of growth or malnutrition which might have resulted if the aromatic amino-acids were not utilised. Furthermore when scorbutic guinea-pigs are given parahydroxyphenyl pyruvic acid, it is metabolized to a considerable extent irrespective of the degree of ascorbic acid deficiency.

The position is not, therefore, clear as to

what part, if any, vitamin C plays in the metabolism of certain amino-acids. The development of an abnormal tyrosine metabolism in the scorbutic state would raise the problem of the metabolism of other essential amino-acids, with their powerful influence on the integrity of the hepatic parenchyma.

From the clinical aspect the solution of this problem would be extremely difficult. The nature of the diet itself, with its poor amino-acid content could provide a mechanism. The associated severe anaemia may play its part or a combination of factors may be responsible.

Finally, permanent parenchymal damage may interfere with the interpretation of the liver function tests following therapy.

No matter the cause, in those cases where daily urobilinogen studies were performed, all urinary levels slowly dropped on bed rest alone, some to well within normal limits. The faecal urobilinogen appeared to reflect the extent and degree of haematoma formation. Case 6 with apparently normal liver function tests revealed liver dysfunction by a marked rise of urinary urobilinogen when fresh haematomata formed. Case 8's urinary urobilinogen remained just above normal despite a gross increase in the faecal fraction with fresh haematoma formation. The colloidal gold reaction was grossly abnormal however. Cases 2 and 23 were the only cases with elevated serum bilirubin levels and the former had bile in the urine, for the first two days after

admission. High icteric indices were frequently found in association with a normal serum bilirubin.

To differentiate anaemia from vitamin C deficiency per se, as the cause, would be impossible. With regard to the serum proteins an increase in the albumen level with a decrease in the globulin fraction following treatment occurred in most of the cases followed up. In these cases however, improved liver function from the recovering anaemia could have been the mechanism. It is of interest to mention here that the iron-binding capacity of the plasma likewise improved with treatment. Liver function, as measured by the turbidity reactions however, was not significantly altered.

Nevertheless the table shows that some of those with the grossest abnormal functions were not anaemic, and these functions did not improve when ascorbic acid only was added to the diet.

Consequently the final conclusion that one may draw from these results is that the probable primary mechanism is the deficiency of essential amino acids in the original diet with the possibility that anaemia may play a contributory part.

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3. Evidence of other nutritional deficiencies.

That avitaminosis is usually multiple is well known. KEKWICK et al (1947) mention this as a possible cause of ill health in their patients. Some have suggested that the full syndrome of scurvy depends in large measure on the presence of associated vitamin deficiencies [CRANDON et al (1940)] Reference has been made to the close similarity of certain scorbutic features to vitamin A deficiency [SHAPIRO (1942), CRANDON et al (1940), MAYER and KREHL (1948)]. VILTER et al (1946) found that seven of their nineteen cases had mild peripheral neuritis, four fiery red glossitis of niacin deficiency and two the magenta-coloured glossitis and cheilosis of ariboflavinosis. McMILLAN and INGLIS (1944) in their large series of fifty-three cases recognised multiple neuritis in three males, and pellagra in two males and one female. FOX and DANGERFIELD (1940)(twenty-eight cases) and RALLI and SHERRY (1941)(twenty-seven cases) make no mention of associated avitaminoses. The nature of the diet in the Bantu in this series apparently supplied enough vitamin B complex in his bread (approximately 90% mill), and the liberal quantities of his mealie meal porridge. The small dry bean, which is often eaten, has a high vitamin B complex content but no vitamin C [EDDY and DALLDORF (1941)].

There was an almost surprising lack of other vitamin deficiencies. Careful search was made for features suggestive of these. In no case were there any signs to suggest

peripheral neuritis. One case of mild scurvy with no anaemia (Case 7) was admitted exhibiting marked general weakness, mental dullness, disorientation. In addition corneal vascularisation, the magenta-coloured tongue and the nasal filiform sebaceous excresences or "sharkskin" nose of ariboflavinosis [SPIES et al (1939)] and the "crazy paving" dermatitis of pellagra, were present. All these features responded promptly to vitamin B complex added to the basic mealie meal diet. Scorbutic gums and perifollicular petechiae remained.

No other coincident vitamin deficiency was noted in the personal series. In fact, one was impressed by their well nourished appearance.

In the remaining nineteen cases of the total series one had slight cheilosis and a "sharkskin" nose of ariboflavinosis, with some slight "pavement" skin dermatitis of the shins of early pellagra. Two others had this last mentioned feature, one with a "sharkskin" nose and the other with no other deficiency features apart from his scurvy. Consequently of the total series of thirty-two cases only four had any manifestations of associated vitamin deficiencies and apart from the first case mentioned, they were mild in each case.

The first case mentioned above had been without work for three weeks and consequently without nutriment before admission. Another was so anaemic (8% packed cell volume) that he had been confined to bed in the quarters at the factory and his diet depended on the generosity of his fellows. The re-

maining two stated that in the preceding two weeks their painful gums prohibited more than a morsel of bread a day. One might conclude from this that the staple mealie meal, bread diet of the Bantu here is sufficient to protect them from vitamin B complex deficiencies.

In one of the four cases mentioned above and in two others of the personal series a smooth atrophic tongue failed to respond to vitamin B complex and responded only after the anaemia responded. All three had histamine-fast achlorhydria.

In the personal series, in an attempt to achieve greater accuracy with regard to the specificity of various scorbutic features, vitamin B complex parenterally ("Bejectal", "Plebex") was given in large doses over several days during some period of the control stage to ensure that no undetectable B complex vitamin deficiency was playing a part.

Table 2.

INCIDENCE OF THE MORE COMMON CLINICAL FEATURES

Clinical Feature	Personal Series (13 Cases)	% Incidence	Remainder (19 Cases)	% Incidence
SKIN MANIFESTATIONS				
Follicular Hyperkeratosis	13	100	16	84.3
*Perifollicular Petechiae	2			
GUM LESIONS				
1. Swelling	13	100	19	100
2. Pain	11	84.5	14	73.6
3. Foetor	11	84.5	7	36.9
4. Obvious Dental Caries	6	46.1	3	15.8
5. Bleeding	4	30.8	5	26.4
DEEPER HAEMORRHAGIC MANIFESTATIONS				
1. Pain in the legs	11	84.5	16	84.3
2. Pain in the arms	2	15.4	8	42.1
3. Haematomata (legs)	9	69.3	17	89.5
4. Haematomata (arms)	2	15.4	7	36.9
5. *Ecchymosis	1		1	
6. Epistaxis	1	7.7	0	
CARDIO-RESPIRATORY SYMPTOMS	6	46.1	8	42.1
OTHER DEFICIENCIES	2		2	

* In the black skin the recognition of this is difficult.

Summary of the Clinical Syndrome and Complicating Factors of Scurvy.

1. Skin changes, gum changes and deeper haemorrhagic manifestations are the basic features of the scorbutic syndrome, and appear in that order as the disease progresses.

2. The first skin change is follicular hyperkeratosis followed by perifollicular petechiae. From this stage on wound healing is delayed.

3. Gum changes vary from no obvious change except localised interdental gingival hypertrophy, which may be missed if not diligently sought for, through all degrees to large granulomatous excrescences, extending on to the palate.

4. Deeper haemorrhagic features manifest themselves as ecchymosis, which is not easy to see in the Bantu, to large intramuscular and less often subperiosteal haematomata. The neighbouring joint usually shows an effusion while the overlying skin, and if in the calf, the ankle, may show oedema. Other cardinal features of inflammation are present in the swelling. Bleeding from the mucous membranes does occur but is not often seen. These haemorrhages give rise to the crippling pain associated with the disease.

5. Other features noted have been pallor, drop in blood pressure, water retention, loss of weight and pyrexia.

6. The type of diet on which scurvy develops may lead to other

avitaminoses, and liver dysfunction. It was felt that superadded infections played no part in the formation of the scorbutic syndrome.

7. By placing the patients on the same diet on which the disease developed and adding only ascorbic acid, the specificity of these scorbutic features was proved. The grosser gum changes responded the quickest although some improvement was noticed merely by clearing the secondary infection. The haematomata began to absorb on bed rest alone but residual thickening could still be appreciated four to six weeks after ascorbic acid. Follicular hyperkeratosis, being the first to appear, was the last to go, often taking as long as two months to do so.

8. Finally, although it may not be proven that follicular hyperkeratosis is only due to vitamin C deficiency, it would appear, on the other hand, that the diagnosis of scurvy in its absence is not warranted.

C H A P T E R IV.

THE DIAGNOSIS OF VITAMIN C SUBNUTRITION.

THE CONCEPT OF SUBCLINICAL SCURVY.

The Diagnosis of Vitamin C Subnutrition.

The haemorrhagic tendency that occurs in scurvy and the fact that ascorbic acid is a powerful reducer of the redox dye, 2:6 dichlorophenolindophenol, has led to numerous technical and chemical tests for the detection of vitamin C subnutrition. The details of the methods are discussed in Appendix I.

A. Tests dependent upon the Bleeding tendency.

1. Capillary Fragility Test.

In the personal series of thirteen cases this test was carried out on each case. In no case could increased capillary fragility be demonstrated by the appearance of petechiae. However in four cases - all with a severe degree of scurvy - large haematomata appeared down the forearm. Our first experience of this caused considerable surprise but on consulting the literature it was evident that this was no new phenomenon, experimentally in guinea-pigs [DALLDORF (1931)] or in humans [McMILLAN and INGLIS (1944)].

The absence of any positive results may have been from the difficulty in interpretation due to the black colour of the skin as other workers on Bantus have not obtained positive results [FOX (1940)(1941) and KEKWICK et al (1947)].

Having this in mind the last mentioned authors used the

inside of the lip and applied negative pressure with inconclusive findings.

The literature abounds with articles both in favour and against the use of the capillary fragility test for the diagnosis of vitamin C subnutrition. [HESS (1920), KENNEY et al (1939), RALLI and SHERRY (1941), BROCK (1942), SHULTZER (1933), GOTHLIN (1937), JENNINGS et al (1938), CRANDON et al (1940), KEKWICK et al (1947), LIEBMAN et al (1938), SHAFAR (1949), ABBASSY (1935), FOX (1941), UNGLEY (1938), McMILLAN and INGLIS (1944), MUNRO et al (1947). Apparently it is not only in Bantus that severe scurvy gives rise often to negative results [SHULTZER (1933), McMILLAN and INGLIS (1944)], the latter authors even using different methods and noting whether any change occurred after vitamin C therapy. In forty patients it was positive before, and in only twenty-one was it negative after therapy. Mild scurvy experimentally produced does not give positive results [CRANDON et al (1940), PETERS et al (1948). When one considers the variety of conditions that may give positive results [HOLLAND et al (1947), LIEBMAN et al (1938)], one need not be surprised at the conflicting opinions as to its value and the different techniques that have been suggested to ensure greater accuracy. GOTHLIN (1937) who popularized the test is now diffident and stresses its use as a "screening" test in mass experiments, but adds

that the petechiae resulting, must be less with subsequent testing after vitamin C therapy. Daily variations, and different responses on the two arms of a normal subject have been noted [GREENE (1934)]. Furthermore some cases of scurvy showing positive results may clear while the scurvy is progressing [SHULTZER (1933), UNGLEY (1938)]. Consequently even a retrospective diagnosis of scurvy cannot be made with certainty should the petechiae lessen after vitamin C therapy.

In considering the larger reviews on the subject [ABT and FARMER (1938), LIEBMAN et al (1938)] and the values in controlled experiments in humans [CRANDON et al (1940)], RALLI and SHERRY (1941) agreed with other authors that all these capillary fragility tests were of little value in diagnosing vitamin C deficiency.

2. Bleeding time.

In all cases of this series on which this test was performed, normal results were obtained. The only abnormal findings in the literature consulted are a bleeding time of 8½ minutes in one case reported by JENNINGS and GLAZEBROOK (1938) and another of 35 minutes by HATHERLEY (1947) but this was a most atypical case, if scurvy was the diagnosis. The weight of evidence [McMILLAN and INGLIS (1944),

NINENSON and COHEN (1937), SHATTUCK (1928) and WOOD (1935)] suggests that a normal bleeding time accompanies scurvy.

3. Coagulation time.

All cases (sixteen) of the thirty-two cases studied in this respect showed a normal coagulation time. This agrees well with results of other workers on this aspect of scurvy [WOOD (1935), NINENSON and COHEN (1937), JENNINGS and GLAZEBROOK (1938), McMILLAN and INGLIS (1944)].

4. Platelet Count.

Likewise, normal values were recorded as did WOOD (1935), YOUNG (1938), SHATTUCK (1928) and McMILLAN and INGLIS (1944).

5. Prothrombin times.

VILTER et al (1946) found normal prothrombin levels in seven of their eleven patients. Of the seven cases in this series in whom this was investigated, only two showed slightly longer times than the controls. It is doubtful whether any significance can be attached to these two cases as liver dysfunction, as determined by chemical methods, was present.

B. Tests dependent on the rapid reduction of the redox dye.

1. Intradermal dye test.

This test was attempted on one patient but owing to the black colour of the skin no colour change could be accurately assessed. This together with the unsatisfactory reports in the literature made one abandon the test.

These unsatisfactory reports are stressed in recent [HOLLAND et al (1947)] and older reviews [GOLDSMITH et al (1939), FOX (1940), RALLI and SHERRY (1941)]. WRIGHT and MacLENATHEN (1939) found variations in different parts of the forearms even. A normal result has been found in severe scurvy [JENNINGS and GLAZEBROOK (1938)]. When one considers that vitamin C is only one of the reducing substances in the skin, with the vasomotor state, the thickness of the skin and the temperature all helping to produce variations, one cannot but expect that this would not be a helpful test [FOX (1940), SHAFAR (1949)].

2. Plasma Ascorbic Acid.

This has been one of the most widely used tests to determine the vitamin C status, yet as far as single readings are concerned, it is one of the most unreliable. Our findings, i.e. a case of severe scurvy having a plasma level of 0.35 mg. %, agreed well with ABT and FARMER's (1938) findings that levels of 0.4 and 0.5 mg.%

occur in active scurvy. Furthermore FOX and DANGERFIELD (1940) report that their cases of scurvy did not invariably have lower values for plasma ascorbic acid than individuals who appeared to be in an excellent state of health. BUTLER and CUSHMAN (1940) noted the rapid changes in plasma ascorbic acid concentrations following the addition to, or withdrawal of ascorbic acid from the diet. McMILLAN and INGLIS (1944) on twenty-two cases of scurvy, noted that, although all had low values (up to 0.3 mg.%), an inconstant relation existed between these figures and the clinical picture. PORTNOY and WILKINSON (1938) warn that the plasma level is subject to sudden fluctuations after a temporary deficiency in the diet. Day to day variations have been reported [BUTLER and CUSHMAN (1940)] and some [FAULKNER and TAYLOR (1938), UNGLEY (1938) and SHAFAR (1949)] maintain that the level in the plasma depends upon the renal threshold.

The finding of a low ascorbic acid level has now been accepted as, in itself, not providing a reliable measure of either vitamin C deficiency or the degree of tissue unsaturation [RALLI and SHERRY (1941), FOX (1940), CRANDON et al (1940), PETERS et al (1948), EDDY (1941). A rapid diffusion of ascorbic acid takes place from the plasma into the leucocytes. Zero or near zero levels have been reported by

many with no clinical signs present [CRANDON et al (1940), PETERS et al (1948), PRUNTY et al (1943)] or so low that it was detected with difficulty in patients in perfect health [FOX (1941)]. In experimental human scurvy [CRANDON et al (1940), PETERS et al (1948)] the plasma level became near-zero or zero after 37to 41 days of a diet totally deficient in vitamin C and remained at that level for 90 to 100 days before the first clinical signs of scurvy became manifest. It is also of interest that in the one series [PETERS et al (1948)] approximately the same level was maintained by those individuals on a supplement of 10 mg. a day; individuals in whom no signs of scurvy developed over a prolonged period (up to fourteen months). Furthermore a supplement of 20 mg. a day produced no rise in the plasma concentration. In those receiving 70 mg. a day, the initial level of 0.5 to 0.6 mg. % was maintained yet when their maintenance was dropped to 50 mg. a day their plasma levels dropped to 0.3 mg. per 100 ml. From these results the conclusion reached by Brock in 1942, some years before these experiments were performed, is well confirmed. He concluded that although the plasma level may be able to distinguish between individuals on different intakes at the higher levels (approximately 40 mg. a day), it would appear to be unreliable for those very individuals in whom the existence of subclinical scurvy is most at issue, i.e.

where the intake is below 40 mg. a day.

Therefore high levels of plasma ascorbic acid have been reported in scurvy and zero or near-zero levels have been reported in individuals in perfect health.

Despite this however, we find some amazing conclusions drawn by some authors. Many of these conclusions, furthermore, have been referred to repeatedly by less critical authors to prove the specificity or non-specificity of certain features in scurvy.

3. Urinary excretion of vitamin C.

The mechanism of vitamin C excretion has been studied in great detail by Friedman, Sherry, Ralli and Rubin (1938) (1940). This excretion, at any plasma concentration, is determined by the plasma concentration, by the rate of glomerular filtration and the rate of tubular reabsorption. It was further observed that the rate of tubular reabsorption was limited by a maximal rate and when the vitamin is presented to the tubules by glomerular filtration at a rate exceeding this, the excess was excreted in the urine. Even at the lowest plasma levels the reabsorption of the vitamin is never complete and there is a minimum amount excreted in the urine

Some workers [FAULKNER and TAYLOR (1938), CRANDON et al (1940)] feel that there is a renal threshold for vitamin

C, although one [CRANDON (1940)] feels that the threshold drops in the totally deficient state and rises again as "saturation" with vitamin C takes place. Other workers however [KELLIE and ZILVA (1939)] feel that no constant threshold exists but that there is rather competition for the ascorbic acid of the blood by the absorptive capacity of the tissues and the excretory function of the kidney. Furthermore renal dysfunction interferes with the excretion, and the vitamin C excretion may fluctuate during the course of the day [SHAFAR (1949)].

Apart from these physiological difficulties there are several technical difficulties. Titration must be rapid to prevent the slower reducing effect of other substances present (e.g. thiosulphates). Furthermore the larger amount of urine that has to be used when low vitamin C levels exist, makes the end point difficult to assess accurately.

In the personal series, seven of the thirteen cases were studied and the vitamin C excretion in each case as measured by the chemical test, was below 10 mg. per day. In one case the levels were followed over twenty consecutive days with results varying from 2.0 mg. to 5.5 mg. per twenty-four hours. Considerable fluctuationsin concentration of the vitamin C excretion were found throughout the twenty-four hours

on urines tested immediately after voiding. In no case studied was there detectable evidence of renal dysfunction. Controls, whose levels were estimated at the same time, with the same reagents and method, agreed well with the given normal figure of above 20 mg. per day.

These universally low results in scurvy are, however, not always seen and this is not surprising in view of the physiological and technical difficulties mentioned above. SHULTZER (1936) records that one of his three cases of scurvy was excreting as much vitamin C (24 mg.per day) as his controls.

On the other hand low excretions may be found in individuals in perfect health [EVELYN et al (1938), ROE and HALL (1939), FOX (1940)]. This, too, then appears to be an unreliable test for the assessment of vitamin C nutrition.

4. "Saturation" test.

Wright's method was used in seven patients of the personal series. In five it took between four and five days of 1,000 mg. intravenously before 400 mgm. was excreted in the urine. In the other two patients after ten days of the above dose, no more than 250 mgm. was obtained. Such a state of "unsaturation" is hard to imagine.

PORTNOY and WILKINSON (1938) prefer this test to all others, but unfortunately it is subject to all the faults of urinary estimation, mentioned above, together with the fact that considerable fluctuations may exist during the course of the day. As the urine is collected for only a short period of the day, i.e. for five hours after the injection is given, this phenomenon may produce considerable differences. From the technical point of view the intravenous injection has to be given very slowly so that the plasma concentration does not rise above the renal threshold.

Both KEKWICK et al (1947) and FOX and DANGERFIELD (1940) found low values in healthy people whereas McMILLAN and INGLIS (1944) in their large series, found no steady relationship between the saturation requirement and the clinical extent of the disease, one of their scorbutics being saturated within the normal length of time. ZILVA (1944) showers very strong criticism on these saturation tests as there are so many different methods of administration, different "test" doses and different criteria for the end point - "arbitrary levels imposed by themselves in accordance with their degree of enthusiasm". He also remarks that no evidence exists that the vitamin C in the tissues of the saturated person is stored for reserve purposes, as results obtained in guinea-pigs point to

the contrary.

Lastly, SHAFAR (1949) points out that a delayed appearance in the urine may occur in renal dysfunction and so interfere with the interpretation of the result.

5. "Tolerance" test.

Here again different doses and routes of administration are used and the same principles for "saturation" exist with their consequent limitations. Unfortunately this particular test has not been widely used. KEKWICK et al (1947) produce a graph to show the high rise and slow fall that is found after large doses of vitamin C supplements have been given over a long period, i.e. a "saturated" state.

6. Ascorbic acid content of the White cell-platelet layer.

In the blood the highest concentration of ascorbic acid is found in the white cell-platelet layer. Normally this is twenty to forty times the concentration of the plasma ascorbic acid, whilst the red cell levels are one to two-and-a-half times greater than the plasma levels [SHAFAR (1949). A rapid diffusion of ascorbic acid takes place from the plasma to the leucocytes, whereas this process occurs at a slow rate into the erythrocytes [SHAFAR (1949)]. That this is the most sensitive test of vitamin C nutrition is adequately demonstrated by the well-controlled experiments on

human volunteers [PETERS et al (1948), CRANDON et al (1940)]. Here the ascorbic acid content of the "buffy" layer became zero only ten to forty days before the appearance of scurvy. Unlike the plasma this level fell slowly. In those volunteers receiving the 10 mg. supplement per day, however, the levels were only 1 mg. per 100 G. more than in the totally deficient group [PETERS et al (1948)]. Some doubt exists [ZILVA (1944), PETERS et al (1948)] as to the accuracy of determination below 2 mg. per 100 G. white cells. It is remarkable that the supplement of 10 mg. a day - sufficient to prevent scurvy - hardly affected the concentration in the plasma and white cells. It is noteworthy also that the white cell layer was the first to become saturated following treatment [BUTLER and CUSHMAN (1940), PETERS et al (1948), CRANDON et al (1940)]

There is no doubt then that this appears to be the most sensitive test for vitamin C subnutrition. However, as RALLI and SHERRY (1941) point out such a test is both tedious and difficult and not readily adaptable for routine laboratory use. It has been shown further, with regard to subclinical scurvy, i.e. if such a condition exists, that even this sensitive test may be zero for six weeks before any deterioration in health occurs and then an asymptomatic clinical sign is all that appears. Furthermore near-zero levels

(2 mg. per 100 G.) may be present without scurvy ever appearing i.e. on individuals receiving 10 mg. supplement of ascorbic acid daily [PETERS et al (1948)]. The Medical Research Council of Britain [PETERS et al (1948)] conclude that a concentration below 2 mg. per 100 G. white cells, especially when confirmed on repeated analysis, indicates severe depletion and supports the diagnosis of scurvy.

Estimations on whole blood are at a disadvantage in that alterations in the white cell count as in leukaemia, leucocytosis and leukopaenia tend to invalidate the readings [BUTLER and CUSHMAN (1940), SHAFAR (1949)]. Haemolysis, also, is a very important factor to be avoided in this method and conflicting evidence regarding the analytical procedures exists in the literature [BUTLER and CUSHMAN (1940)]. Deficiency follows the same trend as the white cell-platelet layer.

Conclusions as to their value.

Of all the tests mentioned several conclusions can be drawn:

1. One must agree with KEKWICK et al (1947) that the ascorbic acid metabolism of the Bantu appears to differ in no way from that of other races.

2. That the ascorbic acid content of the white cell-

platelet layer, despite the difficulty of its estimation is the most sensitive and practical test for vitamin C subnutrition, but it must be remembered that a low level, particularly a single estimation, does not necessarily mean ill-health.

3. That to attribute certain features as being due to vitamin C subnutrition, purely on the basis of one or more of these tests, is both dangerous and unwarranted.

The Conception of Subclinical Scurvy.

In manifest scurvy, apart from the features mentioned in the previous chapter, certain vague and ill-defined symptoms have been described.

Fatigue was the first symptom described by CRANDON et al (1940). There was some decreased ability to perform aerobic work after three months of total deprivation, but no loss in anaerobic work. This occurred prior to the appearance of follicular hyperkeratosis. The occurrence of fatigue as the first feature has not been confirmed by other experimental human studies [RIETSCHEL and MENSCHING (1939), PETERS et al (1948), ANDREW (1949]. ANDREW (1949) complained of fatigue but that was at least a month after the appearance of follicular hyperkeratosis. He found that travelling became an effort and later irritability unwarranted by the provocative incidents was noticed. RALLI and SHERRY (1941), VILTER et al (1946) and SHAFAR (1949) mention general loss of vitality, undue fatigue, disinclination for physical exertion, anorexia, mental depression and negativism. No such subjective symptoms were experienced by the volunteers in the Medical Research Council report [PETERS et al (1948)] but abnormal fatigue tests were reported after the first sign of scurvy appeared.

McMILLAN and INGLIS (1944) make no reference to these subjective features at all amongst their large series

of fifty-three cases. FOX (1941) does not mention these symptoms but remarks on the characteristic languor and incapacity for work of frank scurvy.

In this series generalised weakness was complained of in seven of the thirty-two cases, but in all, except one, gross anaemia could have accounted for it. Multiple avitaminosis was present in the non-anaemic patient. This patient (Case 7) had great difficulty in sitting upright in bed. This weakness rapidly disappeared three days after the addition of vitamin B complex only to the diet on which the disease developed. Mental dullness was often difficult to assess due to language difficulty, but co-operation on the patient's part was never lacking. One patient was irritated by the strict routine in collecting stools and urines for pigment studies, but it was felt that this was not an unwarranted provocation. After vitamin C therapy, however, the patients were strikingly more cheerful. The feeling was that this response was no more significant than that of any patient responding to an unpleasant illness.

Nevertheless it is possibly these vague symptoms which first gave rise to the concept of subclinical scurvy.

EDDY and DALLDORF (1941) feel that two varieties of subclinical scurvy exist, one an "asymptomatic form of deficiency" and the other a mild atypical form of symptomatic scurvy.

The latter masquerades as rheumatism, gingivitis, purpura, haemophilia and osteomyelitis. They add, however, that a definite diagnosis can only be made by cure with specific treatment. One feels that this is really atypical scurvy and does not fall into the category of subclinical scurvy to which most refer. By virtue of its definition - "prescorbutic or subclinical" - its existence depends on the chemical or technical tests just mentioned. Before the unreliability of these tests was universally accepted this "unsaturated" or "subscorbutic" state had been blamed by earlier writers for causing a decreased resistance to infection, deterioration of physical and mental efficiency, purpura, anaemia, bleeding in Typhoid fever, and even peptic ulcer and its complications [BOURNE (1938), EDDY and DALLDORF (1941).

EDDY and DALLDORF (1941) point out that it is a natural feature of the avitaminoses in contrast to the major infectious diseases, for example, that all degrees of deficiency, and therefore presumably of ill health, may occur. Although such is very true, it is on the last phrase - the question of presumed ill-health - that the controversy rages. Likewise PARSONS and SMALLWOOD (1935) state that a deficiency of vitamin C does not produce an "all or nothing" response but they admit that proof of the existence of latent scurvy is lacking. BROCK (1942) puts it in another way - "Is the individual, who is consuming the indispensable minimum for the prevention of scurvy only walking on the edge of a precipice from which he can be precipitated suddenly into the chasm of

scurvy by added calls on his reserves, such as infection or very strenuous exertion, or is he, on the other hand, already part way down an incline leading to the chasm of scurvy?". He favours the latter view but states that it cannot be proved.

As SMITH (1938) remarks, "With increased study as so often is the case the problem has become more complicated than it then seemed", whereas RALLI and SHERRY (1941) mention that "the advances in our understanding of this condition have come through improved technical and chemical methods" and they go on to mention that the question of the existence of the prescorbutic state and those degrees of vitamin C deficiency which were not profound enough to cause the clinical signs of scurvy can only be diagnosed by the use of chemical methods. More recently ZILVA (1944) refers to the reckless application of "these improved chemical methods" to diagnose hypovitaminosis C "when there is as yet no definite evidence to show at what stage 'unsaturation' may have an unfavourable influence on health, but what evidence there is, suggests that it is likely to be in the zone immediately preceding the frank manifestations of the disease."

The wisdom of Zilva's remark could be well applied to the conclusions drawn by CROFT and SNORF (1939) some years earlier - "In no case did we find spontaneous petechiae or subcutaneous or submucous haemorrhages and other than a high incidence of caries and gingival infections, no other suspicious symptoms of scurvy were observed. We are unable to

account for the absence of clinical scurvy in people with a definite depletion of cevitamic acid extending over prolonged periods. Because of this we are led to wonder if some other factor than cevitamic acid, as revealed by the test of FARMER and ABT (1936) may be involved in the production of clinical scurvy in the adult." Another remark in their work is worth quoting. "It is possible that synthetic vitamin C such as we used is not the true anti-anaemic factor in scorbutic anaemia for the above mentioned authors [DUNLOP and SCARBOROUGH (1935)] used orange juice". As can be seen no patient had signs or symptoms of scurvy, yet the anaemia present is called "scorbutic" anaemia. Furthermore the authors they quote used pure ascorbic acid. Their group consisted of a hundred hospital patients with peptic ulcers, cirrhosis of the liver, infections, etc. More likely causes for the "scorbutic" anaemia were present. The plasma levels of ascorbic acid referred to were between 0.1 and 0.2 mgms.%. Another author to whom others refer when they wish to counter the suggestion that there is a specific anaemia due to ascorbic acid lack is LOZNER (1941) who concludes - "Haemoglobin regeneration may occur in the absence of reduced ascorbic acid from the body by the chemical test". CARTWRIGHT (1947) wisely criticizes this remark by noting that only the unreliable plasma ascorbic acid determinations were made and reminds readers that one of Lozner's five cases was complicated by a bleeding ulcer, another by alcoholic pellagra and a third was a woman with a history of nine pregnancies, achylia and a urinary tract infection. Conclusions such as the above, based on these

"improved technical and chemical methods" with reference to other so-called "subscorbutic" features, abound in the literature, and are pounced upon by uncritical authors to propound the ill-effects of the subclinical scorbutic or "prescorbutic" state.

This state, it must be emphasized, is dependent upon chemical and technical tests, which have just been shown to be unreliable. More definite criteria for a state of "unsaturation" would be, not only these abnormal tests, but a known intake of a diet low in vitamin C. Furthermore this low intake must have been very low (possibly in the region of, or below 10 mg. per day) and must have been very prolonged.

FOX et al (1940)(1941) having at their disposal about a 1000 Bantu mine labourers with plasma levels so low that "they could be detected only with difficulty" and other criteria necessary for a state of "unsaturation", set about this problem.

Briefly their findings were:-

1. There were no signs of impaired health.
2. Increased capillary fragility could not be demonstrated, even in frank scurvy.
3. No anaemia resulted.
4. The gums were healthy throughout [GOTTLICH (1940)].
5. There was no indication that resistance to infection had diminished.
6. There was no impairment of the healing of wounds.

7. This deficient group was no worse from exercise point of view , tested with regard to strength (Shot Putt), skill (100 yards sprint) and stamina (one mile race) than another group receiving an additional ration of orange juice [JOKL et al (1940)].

8. The development of scurvy depended on extremely severe and prolonged deprivation of vitamin C without precipitating factors. Despite the same long continued low daily intake of vitamin C only twelve of their large group developed scurvy.

Likewise KEKWICK et al (1947) found that the incidence of scurvy was low amongst their East African Bantus despite the fact that plasma levels were well below those regarded as satisfactory for Europeans. The average daily intake in this group was at or below 15 mg. per day.

JENNINGS and GLAZEBROOK (1938) comment from one of their two cases on the length of time during which scurvy may remain latent. "For forty years, at least, this man had lived on a diet grossly deficient in vitamin C" yet his symptoms were of recent duration.

Confirmation of these observations is forthcoming from the well-controlled experimental work on human volunteers. Mensching [RIETSCHEL and MENSCHING (1939)] showed no signs or symptoms after a hundred days on a scorbutogenic diet. In the other two series [CRANDON et al (1940), PETERS et al (1948)]

about a hundred days elapsed between the disappearance of ascorbic acid from the plasma and the first clinical sign of scurvy (follicular hyperkeratosis). During this period ten cases showed no subjective sensations, abnormal fatigue tests or decreased resistance to infection. Crandon, however, as mentioned already, did show decreased ability to perform aerobic work, but no other complaints were manifest. Despite a loss of 6,000 ml. of blood by venesection, no anaemia occurred. Far from resistance to infection decreasing, he was never more free from upper respiratory infection.

From all these studies then, only one feature in only one patient emerged prior to the onset of follicular hyperkeratosis. This feature was decreased ability to perform aerobic work and then, after three months of a totally deficient diet and after two months of a plasma level of zero. During this time no interference with anaerobic work was discovered. Apart from this, then, no evidence is forthcoming, that there are any early symptoms of scurvy or the subclinical state until well after the first clinical signs appear. In the absence of the characteristic clinical signs the diagnosis of scurvy, or ill-effects due to vitamin C deficiency, on the basis of chemical or technical tests is not warranted. The only presumption that may emerge on this evidence is that of FOX et al's (1940). They state that an individual with a low reserve of the vitamin may develop scurvy somewhat earlier than one whose reserves are well supplied, i.e. a "potential" scorbutic.

If one discards the presence of ill-health in the pre-scorbutic state, has one to adopt the "all or nothing" or "precipice" theory of the onset of scurvy, because, as EDDY and DALLDORF (1941) point out all degrees of deficiency must occur?

With the fore-mentioned facts this is not necessary. The slow, insidious onset of scurvy, dependent upon a very prolonged and almost total absence of vitamin C from the diet, has been stressed already. The human experimental work shows further that the progress of scurvy, once it has announced its presence, is also slow. One must, however, make provision for a precipitating cause, such as infection, lessening the long time intervals between the appearance of each new feature. The first sign is follicular hyperkeratosis and as WILTSHIRE (1919) pointed out, the patients do not notice is appearance, let alone complain of it. The same may be said of perifollicular haemorrhages. In fact, in most of the larger series, stress has been laid on the appearance of pain, due to deeper haemorrhagic manifestations, as the first symptom. The onset and progress then of scurvy is so insidious that a severe stage of the disease is present before the patient is subjectively aware of it. The insidious nature of the disease is probably due to the role played by the stores of vitamin C in the body. Only as these begin to be depleted, do certain features emerge. Possibly as with other avitaminoses, certain structures show the effect of this depletion before others. All the manifestations of scurvy, then, do not appear

in one fell swoop so the "precipice" becomes a gradual downhill slope. The grade or steepness of this slope would depend in large measure on the adequacy or inadequacy of the supply of vitamin C either from the diet or from the body stores and the presence or absence of some complicating factor such as infection.

Summary and Conclusions, on the Diagnosis of Scurvy.

1. The weight of evidence appears to disfavour the conception that ill effects occur in the "pre-scorbutic" or "subclinical scurvy" state.

2. Chemical and technical tests, unfortunately, are too unreliable to be of any value in confirming the diagnosis of scurvy. Accuracy of this diagnosis, as with any deficiency state, is proved, as GOLDEN (1941) points out, by the patient's response to specific vitamin therapy. When such a response occurs, the zero level of the blood [ZILVA (1944)] only offers additional support to the diagnosis.

3. The value of follicular hyperkeratosis, then cannot be overstressed. Confirmation of a possible scorbutic lesion by the appearance of the gums is unreliable. The edentulous show no changes, while those with healthy teeth may also show no changes. Pyorrhoea alveolaris is a common complaint. It would be fruitless giving vitamin C, if such a deficiency was not the underlying cause. On the other hand, complete resolution will not occur, if it is the underlying basis, unless vitamin C is given. Dentists would do well to remember this simple clinical sign in such cases.

This was borne out in one case of this series. A dentist referred a fungating malignant neoplasm of the palate and alveolar margin to the Ear, Nose and Throat outpatient department. The "neoplasm" melted away on vitamin C therapy. Follicular hyperkeratosis was present.

In the Surgical outpatient department a biopsy was taken of an epitheliomatous ulcer of the ankle. The report of chronic inflammation associated with his severe anaemia in this Bantu made sickle cell anaemia the admission diagnosis. Follicular hyperkeratosis was present.

Another diagnosis on the case with pain in the limbs and chancroid was "Soft sore; Arthritis ? Gonococcal and Anaemia unexplained". Follicular hyperkeratosis was also noted.

A general practitioner sent in a case with a scorbutic haematoma of the calf, diagnosed as a sarcoma.

Four cases were diagnosed as cellulitis with underlying osteitis. Only after several negative X-Ray examinations was the haematoma of scurvy considered. In all these cases follicular hyperkeratosis was present.

Again this useful sign can be applied to surgical wounds. Delayed healing occurs only after this sign has appeared.

4. It will be seen, then, that in the words of RALLI and SHERRY (1941) and others [FOX (1941), ABT and FARMER (1938) and GOLDEN (1941)] the diagnosis of scurvy depends on sound clinical observation and that the specificity of any feature and the final proof of the diagnosis depends upon the patient's response to pure ascorbic acid therapy.

CHAPTER V.

THE ANAEMIA IN SCURVY.

REVIEW OF THE LITERATURE.

Review of the Literature.

The earlier writers of nearly three centuries ago noted pallor in scurvy. TURNER (1911) quotes Syndenham who in 1680 wrote: " .... the legs are sometimes swollen, sometimes emaciated and always have on them livid, lead coloured, buff or violet spots, the colour of the face is usually pale sallow."

In the times of Barlow during the last century pallor was mentioned and he describes the "profound anaemia" [PARSONS et al (1933)] asdid HESS (1920) but other authors [ROHMER and BUNDSHELDER (1932)] ascribe the pallor to circulatory changes.

In South African literature MACVICAR (1906), exemplifying the excellent clinical observation of the earlier workers describes the "characteristic greyish anaemic appearance" of his Bantu patients. Five years later TURNER (1911) remarks that although the complexion was difficult to assess in Bantus "the inside of the lips and gums are pale from anaemia" and a progressive anaemia develops.

ARON (1939) comments that during the period of food shortage in 1921 to 1923 an extraordinary increase in the number of cases admitted to the Children's Hospital Breslau for severe anaemia coincided with a "hitherto unknown" number of cases of scurvy. Fifteen of these cases of anaemia were studied carefully clinically and by X-Ray for signs of scurvy

and such were found in seven. He then quotes ROHMER and BUNDSHELDER (1932) as finding anaemia in seven of fifteen cases of scurvy. These authors also investigated twenty-two anaemic infants and found that in six patients, iron had no effect but with vitamin C a prompt cure resulted. From these findings they are prepared to conclude that vitamin C has some effect on the metabolism of iron! PARSONS and HAWKSLEY (1933) offered a more simple and adequate explanation in that more than one factor, e.g. iron and/or vitamin C may be lacking in some cases.

Just prior to this, some of the first reports of the haematological features of the anaemia in scorbutic guinea-pigs were published. MEYER and McCORMICK (1928) and later, METTIER and CHEW (1932) showed that anaemia was of regular incidence in guinea-pigs on a scorbutogenic diet. The former authors felt that increased blood destruction was the mechanism, whilst the latter, also observing the increased reticulocyte levels, did not agree. In a very extensive study on the bone marrow changes, before and at the peak of the reticulocyte crises after orange juice was given, METTIER and CHEW (1932) drew the conclusion that retarded maturation of the red blood cells occurred in experimentally induced scurvy in the guinea-pig. These authors did not feel that the extent of the haemorrhages found at port-mortem was sufficient to account for a post haemorrhagic mechanism for the anaemia. The fact that from three to four days prior to death increasing numbers of immature cells appeared in both the peripheral

blood and bone marrow suggested that failure to maintain a normal number of erythrocytes in the circulation had resulted in a relative, but inadequate, replacement by a release of cells at a relatively younger stage of maturation. Meyer and McCormick together with Mettier and Chew induced prompt responses when orange juice was fed.

With regard to adult scurvy at this time, SHATTUCK (1928) reported thirteen cases on whom blood counts were performed. The lowest red cell count recorded was 2.4 million per cu. mm. Five cases had a red cell count below 3 million cells per cu. mm., four were between three and four million, the highest count of the remaining being 4.5 million cells per cu. mm. One case died of congestive cardiac failure and the others responded to an antiscorbutic diet with orange juice.

Further information was gained on this anaemia by METTIER, MINOT and TOWNSEND (1930) on studying eight anaemic cases in nine adult scorbutics. They showed the world that this anaemia could be very severe. Both iron and liver, which was prepared in such a way that it was free from vitamin C, had no effect whilst the patients were on a control diet of boiled milk and white flour biscuits. With only orange juice added to this diet a reticulocyte response with complete recovery occurred. These investigators even showed that on this scurvy producing diet and bed rest an actual progression of the anaemia occurred. The feeding of

fresh raw liver with its high vitamin C content in one and a well balanced diet in another, was sufficient to halt this progress and cause complete regeneration.

Although this as a controlled experiment was excellent, it had not yet proved the specificity of the anaemia of scurvy. Orange juice contains iron and some vitamin B complex as well as vitamin C.

The year after pure ascorbic acid was isolated, JANET VAUGHAN (1934) took up the challenge. She kept an old man of 71 years, with scurvy and a haemoglobin of 56%, on a scorbutogenic diet for twelve days, during which his anaemia progressed. On 50 mg. ascorbic acid daily a "striking" result followed. Furthermore she noted that ascorbic acid had no effect on other anaemias unless they were due to scurvy.

The mean cell diameter in her case was 7.05 u which agreed with METTIER et al's (1930) view that the anaemia was normocytic and normochromic. She recorded too an uneven reticulocyte count "presumably due to the stimulating effect of repeated subcutaneous haemorrhages".

This fact, i.e. that a considerable increase in the reticulated erythrocytes may be present in the severe stage of untreated scurvy was noted the following year by DUNLOP and SCARBOROUGH (1935) in one of their two cases of scorbutic anaemia. This reticulocyte count increased

considerably on adding 60 mgm. of ascorbic acid, which led to rapid and complete cure of the anaemia. Their basis of control was the best thus far as the patients were placed on exactly the same diet on which they developed the disease, a vitamin C free, iron poor diet, and observed for three to seven days prior to treatment. They felt that the response could not be attributed entirely to the cessation of haemorrhages since these had never been severe and the improvement in the red cell count, parallel with the haemoglobin, was too rapid on this iron poor diet to be due to this and not due to the ascorbic acid. Their first case, a severe scurvy with severe anaemia, after seventeen days of 60 mg. ascorbic acid daily had increased his red cell count by two million and his haemoglobin by 30%. Ascorbic acid was then stopped and on the deficient diet alone regeneration continued. They showed from this that ascorbic acid is capable of being stored and exerting its beneficial effects for long periods.

With regard to the adult, then, at this time, scurvy appeared to be associated with an anaemia curable with only vitamin C.

This was yet to be shown in infants and guinea-pigs.

PARSONS and HAWKSLEY (1933) had published one of the first cases of anaemia in scorbutic infants on a controlled diet - a diet of milk and nothing else. A full response was obtained when orange juice only was added. Two years later

PARSONS and SMALLWOOD (1935) kept six anaemic scorbutic infants on the same diet on which the disease developed. They felt that they had proved the specificity of the anaemia by the fact that all cases responded both haematologically and clinically to orange juice. It is also noted here that severe scurvy may occur without anaemia but that the long standing cases presented the most outstanding examples of anaemia. In explaining this they remind one that the whole gamut of the scorbutic syndrome is not present in every case of severe scurvy. Mention is made that the anaemia of scurvy is usually normochromic and normocytic, occasionally macrocytic but never microcytic and in this respect comparison with the blood picture of Cretinism is made. They felt from this that the anaemia of scurvy results from a generalised slowing down of erythropoeisis. However with large haemorrhages into the tissues a posthaemorrhagic state may make the picture hypochromic. Infection may also be a factor in increasing the anaemia of infantile scurvy. They believed, then, that more than one factor may be at play in some cases, e.g. "a child suffering from nutritional anaemia may also develop scurvy and the anaemia of scurvy in which case for complete cure vitamin C, as well as iron, will be required".

Three years later PARSONS (1938) reviewed the situation in infants at that time. He felt that the character of the anaemia was similar to that of adults although he felt that it occurred less frequently in scorbutic infants. He contributed two macrocytic and five normocytic

anaemic cases, two of the latter being slightly hypochromic, in a total series of fourteen scorbutic infants to confirm his previous observations. These cases demonstrated the uselessness of iron but a specific effect of ascorbic acid without any other alteration of the diet on which the disease developed. One must assume that by the term "ascorbic acid" the synthetic variety is meant. It would appear, then, that conclusive evidence now exists that vitamin C is essential for the normal maturation of the red dell.

Later in that year, ABT and FARMER (1938) came to a different conclusion on the literature on infantile scurvy prior to 1938 and from results they obtained in treating a scorbutic infant aged ten months. They felt that a generally deficient diet was responsible for the anaemia. When an adequate supply of iron was present, anaemia did not develop experimentally or clinically. "While it is felt by some that the anaemia of vitamin C deficiency may be one of commission, the result of a dysfunction of the red blood cell forming elements, it is probably more often a condition produced by omission, namely of iron, in the generally deficient diet." They agreed with Parsons in stating that anaemia was more common in adults.

The conclusions reached by both Parsons and the last mentioned authors were in a way confirmed by the reports of five cases of scorbutic anaemic infants treated by KENNEY and RAPOPORT (1939). In three severely anaemic cases, pure

crystalline ascorbic acid induced a response. In one of these the red cells reached normal figures but the haemoglobin remained stationary until iron was given as well. Iron was effective in the fourth after ascorbic acid had failed, the fifth case had an initial red cell count of 4.5 million per cu. mm. Unfortunately no details are given as to the type of diet employed or length of the control period. It is of interest to note that these authors noted an elevated reticulocyte count prior to treatment in the more severe cases.

About this time more experimental reports were coming in. ABT and FARMER (1938) had noted that in young guinea-pigs made scorbutic, no anaemia developed if adequate iron was present in the diet. On the other hand METTIER (1938) could still produce anaemia despite iron supplements in young adult guinea-pigs, but scurvy and anaemia took twenty to forty days to develop in this group whereas in the other group without an iron supplement, scorbutic anaemia was noticed in fourteen to thirty days. The anaemia in both groups, however, was similar and with progression a coincidental rise in reticulocytes occurred. Orange juice produced a complete cure.

The following year the position with regard to scorbutic anaemia in the guinea-pig and its relation to pure ascorbic acid was clarified somewhat by ARON (1939) and SIGAL (1939).

Aron showed a difference between young guinea-pigs of 200 to 300 G. weight and older guinea-pigs of 450 G or more, a difference noted earlier by METTIER and CHEW (1932). Aron, maintaining these guinea-pigs on a scorbutogenic diet for a period longer than the life cycle of the red blood cell, showed that anaemia did not develop whilst a supplement of pure ascorbic acid was ingested. When this supplement was withdrawn all guinea-pigs over 450 G weight developed anaemia and scurvy within twenty days and a supplement of iron did not prevent this. Although younger lighter animals could become anaemic also, they usually succumbed before a distinct anaemia developed. Ascorbic acid by injection or orally cured this anaemia provided not more than a third of the haemoglobin or 25% of the body weight was lost. When cure resulted, haemoglobin regeneration occurred to completion faster and long before the rise in body weight or repair of other body tissues. Unfortunately not enough details are given of the more severe cases. Of six animals four succumbed within three to six days. On the fourth day of the two surviving, germinated oats were given. It would be unreasonable to draw the conclusion that ascorbic acid had failed as METTIER and CHEW (1932) obtained the peak of the reticulocyte response in their cases between the fifth and seventh day.

SIGAL (1939) used guinea-pigs of 600 G weight on a scorbutogenic diet supplemented further with yeast and cod liver oil. On this diet alone and in those getting a supplement of 0.5 mg. ascorbic acid a day as well, scurvy

developed in ten to fifteen days followed by anaemia in ten to twenty-one days. There was a parallel decrease in haemoglobin and the number of red blood cells with marked anisocytosis, poikilocytosis and nucleated red cells in the peripheral smear, terminally. Where, however, a supplement of 3.0 mg. of ascorbic acid was ingested, no anaemia and no scurvy developed.

This rather convincing work not only illustrates the specificity of ascorbic acid in confirming the results of earlier workers, but also shows that the anaemia becomes severe in the later stages of vitamin C deficiency as well as providing a "minimal protective dose" of less than 3 mg. ascorbic acid against scurvy in the guinea-pig.

The latest report to hand, however, [BUDTZ-OLSEN (1950)] has confused the issue again. No anaemia has developed before death in some adult guinea-pigs fed a scorbutogenic diet supplemented with iron parenterally, a diet on which no weight loss occurred yet scurvy became fully developed.

In the meantime isolated case reports on adult scurvy had been issued and various opinions passed and conclusions drawn.

SHULTZER (1936)(1937) reports the occurrence of slight anaemia in two of four cases of scurvy. He showed that scorbutics put to bed on a vitamin C free diet showed marked improvement in their clinical features, which recurred

when the patients were allowed up. These subsequently disappeared after three to four days of intravenous ascorbic acid. Only one case had achlorhydria.

NINENSON and COHEN (1937) described the severe haematological picture in a scorbutic female aged 60 years. The anaemia had a high colour index with elevated reticulocytes, leucopaenia, normal bleeding and coagulation times and a normal clot retraction. No details are given of the diet during the control period but a rapid response occurred with orange juice and "cevitamic acid" given together.

YOUNG (1938) showed that severe anaemia in adult scurvy was not confined to the arteriosclerotic age in reporting a young recluse of 25 years with a haemoglobin level of 20%. A two hundred and fifty ml. blood transfusion caused very little change in the red cell count but rapid regeneration took place on a full mixed diet with iron and marmite added.

The appearance of ecchymoses and haematomata in an edentulous patient led to the diagnosis of scurvy in a patient diagnosed for two months as pernicious anaemia, which had failed to respond to liver, iron or blood transfusion and responded damatically to 600 mg. oral ascorbic acid daily. These authors [JENNINGS and GLAZEBROOK (1938) describe a second case in which a vitamin C free, iron free diet was used instead of the hospital diet, used in the first case. They felt that both cases were cured solely as a result of vitamin C

therapy. The first case, with megaloblastic bone marrow, anisocytosis, "anisochromia", achlorhydria and leucopaenia had consumed a vitamin C deficient diet for forty years. His first complaint was increasing dyspnoea on exertion over three to four years, which suggested to them that anaemia had been the earliest feature. The second was orthochromic and normocytic and as the duration of deficiency of vitamin C was shorter they felt that some relation existed between the extent of vitamin C lack in the tissues and the point at which marrow cell maturation was chiefly affected. They stress the curious finding of a high reticulocyte count before treatment in the severe case.

At this time, then, the specific effect of vitamin C on the adult erythron was universally accepted due to the confirmation with pure ascorbic acid of the earlier reports using orange juice.

UNGLEY (1938), however, was one of the first to cast doubt on this specificity. In three cases of bachelor scurvy with anaemia there was a spontaneous reticulocytosis and he was unable to demonstrate a specific effect of vitamin C. A low vitamin C diet (not vitamin C free) was given. How low, it is not stated and the patients were confined to bed. Further analysis shows that one case was given two doses of 50 mg. ascorbic acid at the outset and then kept for a control period of seven days on a low vitamin C diet. Each case had a high reticulocyte count on admission.

Unfortunately Ungley does not report what happened to the clinical features before he gave the large doses of vitamin C.

For the next few years report followed report disclaiming the specific effect of ascorbic acid on erythropoeisis.

MOORE et al (1939) demonstrated that the serum iron curve was higher if ascorbic acid was given with ferric salts by mouth.

CROFT and SNORF (1939) felt that synthetic ascorbic acid was not the anti-anaemic factor in scorbutic anaemia. They arrived at this conclusion by treating six anaemic patients, with low plasma ascorbic acid levels, with 75 to 100 mg. vitamin C daily without success. Furthermore they wondered if some other factor than cevitamic acid, as revealed by the test of FARMER and ABT (1936), may be involved in the production of clinical scurvy in the adult as no signs of scurvy were present in their cases.

FOX (1940) without presenting much evidence remarked that the view that vitamin C has a specific effect on haematopoeisis is now not generally held. In their twenty-eight cases of scurvy FOX and DANGERFIELD (1940) performed blood counts on seven patients, of whom five were anaemic.

In the same year CRANDON et al (1940) showed that despite a loss of some 6,000 ml. of blood in experimental mild scurvy in a human being, no anaemia occurred.

LOZNER (1941) concurred with Croft and Snorf in stating that the universal success with orange juice may be attributed to a factor other than ascorbic acid. He stated that haemoglobin regeneration may occur in the absence of reduced ascorbic acid from the blood by the chemical test. Plasma ascorbic acid levels were estimated. Of five patients with presumptive vitamin C deficiency and moderate anaemia, the haemoglobin of four regenerated spontaneously or in response to iron therapy alone. Rest in bed on a diet having only a trace of vitamin B complex and C was instituted . One case was a female with typical features of idiopathic hypochromic anaemia, chronic cystitis and a history of nine pregnancies. Another case had alcoholic pellagra. The three remaining cases had clinical signs of scurvy. It is of interest to note that in one of these, a gastric ulcer with tarry stools, failed to improve on iron until ascorbic acid was given. In this case and another scorbutic with severe anaemia, elevated reticulocytes were noticed after admission. In the last remaining case venesection of 1600 ml. had to be performed to produce an anaemia. A similar procedure was adopted on the case of Pellagra. It is interesting to compare the two. In the Pellagra, iron caused complete regeneration. In the scorbutic it failed. Oral ascorbic acid (400 mg. daily) likewise failed. After two months of this vitamin B complex and vitamin C deficient diet, it was necessary to give brewers yeast, iron and ascorbic acid before full regeneration occurred.

No note is made whether the patient had developed clinical signs of vitamin B complex deficiency by this time, as a result of this diet.

RALLI and SHERRY (1941) published a long review on Scurvy. Their conclusion, that the absence of vitamin C cannot be considered as solely responsible for the anaemia present, was based on these facts.

Three anaemic scorbutic patients were placed in bed on a diet adequate in all respects except for vitamin C which was present in concentrations below 5 mg. daily. All cases subsequently responded to ascorbic acid. One case during a control period of 52 days prior to treatment showed no further reduction in haemoglobin or number of red blood cells. Ascorbic acid therapy was discontinued on another patient after a response had occurred. After forty-five days whilst still on this scorbutogenic diet no anaemia appeared, although transient petechiae and non-progressive gum changes were noticed on the eighteenth day. Mature erythrocytes, however, have now been shown to survive on the average 120 days [LONDON et al (1950)].

LIU et al (1941) divided sixteen anaemic children in an institution into two groups. One group received iron and the other ascorbic acid. The ascorbic acid treated group failed to respond, which led to their conclusion that the anaemia associated with vitamin C depletion is due to a concomitant iron deficiency. The criteria for vitamin C

deficiency were low plasma levels, an increased capillary fragility and hypertrophied gums. No other bleeding tendency was present. Latent tetany and pulmonary tuberculosis was also noted in the whole group but which children had these features is not clearly stated.

The rapidly disappearing belief that ascorbic acid was necessary for haematopoeisis was checked momentarily by DYKE et al (1942). Three cases diagnosed as pernicious anaemia showed a progressive fall in the red cell count in spite of their usual maintenance dose of liver, in the Spring of the earlier war years. At this time both HARRIS (1942) and FRANCIS and WORMALL (1942) had shown that the diet of the general population during the Winter months was deficient in vitamin C. Without any increase in the dose of liver extract these three cases rapidly improved haematologically and from the general health point of view on 300 mg. ascorbic acid daily. Unfortunately no remark is made whether clinical manifestations of vitamin C deficiency were also present.

It is of interest to note here that ALT et al (1939) found low plasma ascorbic acid levels in both pernicious anaemia and iron deficiency anaemia and attributed this to the achorhydria present. CAYER et al (1946) could not confirm this.

In this respect, ISRAELS (1943) refuses to accept JENNINGS and GLAZEBROOK's (1938) one case as anything but

pernicious anaemia complicated by scurvy. He discusses bone marrow findings in scurvy and contributes three more cases of scorbutic anaemia rapidly improving without any specific anti-anaemic treatment bar vitamin C. Unfortunately no comment is made regarding the diet used.

The largest series of cases of adult scurvy in recent literature was published by McMILLAN and INGLIS (1944). In fifty-three cases of adult scurvy a blood count was performed on forty. In no patient was the blood picture completely normal although in some it was very nearly so - e.g. in two cases there was no oligocythaemia. They stated that anaemia and scurvy need not co-exist in man, and when they do so, the anaemia bears no relation to the extent of the haemorrhages or plasma ascorbic acid content. They summed up the position at that time as:

"(a) Vitamin C lack will not affect blood formation [CRANDON et al (1940)].

(b) Haemoglobin and red cell regeneration with reticulocytosis occurs in scorbutics on a vitamin C free diet [LOZNER (1941), UNGLEY (1938)].

(c) Ascorbic acid has failed to cure an anaemia in experimental scurvy which did react to germinated oats [ARON (1939)].

(d) Vitamin C was necessary in one deficient individual to prevent progress of the anaemia [VAUGHAN (1934)].

(e) Vitamin C is necessary in some deficient individuals before the anaemia will respond to treatment [KENNEY and RAPOPORT (1939), DYKE et al (1942)].

Belief in the need of vitamin C has therfore dwindled and it has been suggested that the anaemia of scurvy is due to lack of other factors [CROFT and SNORF (1939)]. The present experiments confirm points (a), (b) and (e)."

They felt that the anaemia was due to a complex deficiency with vitamin C acting only as an adjuvant, to produce an anaemia usually of moderate degree. To obtain a reticulocytosis, even where the anaemia is severe enough, it would be necessary to supply all deficient factors at once.

The conclusions drawn were:-

1. As no patient showed a drop in either red cell or haemoglobin levels, regeneration however slow must have been going on.

2. Lack of vitamin C alone in the diet did not prevent blood formation.

3. In most individuals its presence accelerated regeneration. For this action little seemed to be required as it was supplied by an ordinary mixed hospital diet (vitamin C content approximately 15 mg./day.).

4. Iron lack was not a serious factor.

5. The anaemias investigated were nutritional in origin. The main causal factor was contained in the vitamin C free diet.

Three cases with megaloblasts in the bone marrow with achlorhydria in two and a low free acid in the third were described. Most cases were normocytic as judged by the mean corpuscular haemoglobin.

Four more cases of scurvy with a high colour index anaemia were reported by GOTTLIEB (1945). All were males between 70 and 80 years of age. Two had very low red cell counts of 1.7 million and 1.35 million per cu. mm. The latter died of congestive cardiac failure five days after admission. The other three responded rapidly to 700 mg. ascorbic acid given intramuscularly daily. He considers that the response was specific despite the fact that the hospital diet was given as well "since the diet did not contain any liver, iron or vitamin B complex; the other recognised haematinics were not present appreciably above minimal amounts."

One of the most detailed and thorough accounts of the anaemia accompanying scurvy in seventeen of nineteen adults is given by VILTER et al (1945)(1946). The two cases not classed as anaemic were mild scorbutics of recent dietary depletion. In addition normal blood counts were found in twelve ambulatory patients with scurvy attending a nutrition

clinic.

Eleven of their anaemic patients with red cell counts varying from 1.74 million to 3.5 million were studied in detail with repeated packed cell volume estimations as well. These patients were placed on a vitamin C free, low vitamin B complex diet. Two patients showed slow but definite clinical and haematological improvement on bed rest alone with clearing of the perifollicular haemorrhages and ecchymoses. Despite this the plasma vitamin C remained at zero. They felt that this response was due to diminished metabolic requirement. In this diet, however, protein in the form of eggs, cheese, potatoes, etc., was present. Another deficient factor may have been supplied.

Nine other cases classed as severely ill did not show any clinical improvement during the observation period which varied from two to twenty-one days depending on the deterioration of the patient's condition. Six cases remained unchanged haematologically whilst in three the blood picture became rapidly worse. All cases responded rapidly to synthetic vitamin C in doses of 500 mg. daily reaching 3.5 million red cells and 12.5 G haemoglobin per 100 ml. or more within three weeks. The ascorbic acid was given intramuscularly in some, and orally in others. In two cases a rise in reticulocytes occurred between the fourth and seventh days followed by a fall, whilst in the other seven the high reticulocyte levels were maintained or rose slightly between the fourth

and seventh days, subsequently falling slowly to normal.

They conclude that vitamin C is essential for normal formation and maintenance of erythrocytes but feel that a patient with severe vitamin C depletion may have no anaemia until additional strain is placed on the bone marrow by a deficiency of extrinsic factor, protein, iron or other unknown factors which may be necessary for normal haematopoeisis. Yet, after the anaemia has developed, the deficiency of the latter factors may not be severe enough to prevent a haematological remission when large amounts of ascorbic acid are given.

They felt that this concept of a multiple deficiency state, where the altered physiology induced by the deficiency of one essential nutrient may increase the need of the cells for other essential nutrients, with many factors besides vitamin C adversely affecting the bone marrow, could explain both the reason why anaemia develops in some and not others and the numerous conflicting reports on scorbutic anaemia which have appeared since 1930.

In the same year further reports on scorbutic infants and children emerged. DOGRAMANCI (1946) reviewed the clinical features of 241 cases of infantile scurvy over the previous ten years and found that in 12% anaemia, usually of a hypochromic type occurred. SCHULZE and MORGAN (1946) felt that no relation between iron and ascorbic acid appeared

to exist in the synthesis of haemoglobin. This statement was based on the fact that children with a low colour index anaemia and borderline ascorbic acid status responded as well to iron therapy alone, as it did when a daily supplement of 100 mg. ascorbic acid was given. The "borderline ascorbic acid status" was assessed on plasma levels between 0.55 and 0.76 mg. per 100 ml., a level that could correspond to an intake of 70 mg. ascorbic acid a day, an intake seven times as great as the minimal protective dose [PETERS et al (1948)]. No clinical signs of scurvy were present.

Although CRANDON et al's experiment (1940) would appear to eradicate the conception that ill health with respect to low haemoglobin levels is a feature of subclinical scurvy, KEKWICK et al (1947) showed disagreement. Of 474 East African Bantus whose average daily intake of vitamin C was in the region of 15 mg., sixteen uncomplicated cases of anaemia with no signs of blood destruction or regeneration were selected. These sixteen were placed on a vitamin free, low protein basal diet with adequate calories and iron for seven to ten days. No change in the anaemia was found. Next, 700 mg. ascorbic acid daily was given. Nine cases showed a response but only three of these reached 100% haemoglobin. The response in six cases was a rise of not less than 18% in the haemoglobin level and a million red cells per cu. mm. Three of these showed a reticulocyte response between the seventh and tenth days. Mention is made that there was a low incidence of scurvy amongst the 474 men

studied but no mention is made as to whether these sixteen men had signs of scurvy or not and no further haematological details are given.

In the same year BARNES (1947) reported a case of severe scurvy with a haemoglobin estimation of 42%. On a strict, sterilized milk diet and an addition of only 20 mg. ascorbic acid - the smallest recorded dose of ascorbic acid used in the treatment of severe scurvy - the patients gums had improved by the fifth day. His haemoglobin was 70% three weeks after the commencement of therapy, in which no other haematinic drugs of any kind had been given. This excellent response to such a small dose in a well controlled experiment lacked only a preliminary period of bed rest prior to ascorbic acid therapy, to determine whether response to bed rest, due to diminished requirement, had played any part. Barnes felt that the anaemia was caused by haemorrhage into the legs, using a bilirubinaemia of 2.1 mgs. per 100 ml. and the rapid recovery without haematinic drugs for support. The blood was "reabsorbed and refashioned".

HATHERLEY (1947) found that a female aged 35 years with gross anaemia, thought to be due to haematemesis from a peptic ulcer, failed to respond to blood transfusion - even as much as thirteen pints - prior to ascorbic acid therapy. There was a dietary history of vitamin C deficiency, no gastro-intestinal lesion was found at laporotomy, no urinary ascorbic acid was found and haemoptysis occurred later. No

mention is made as to the presence of follicular hyperkeratosis but the gums were noted to be healthy. A "dramatic response" occurred to 1000 mgm. vitamin C.

Soon after this, confirmation of Crandon's experiences was given in a more extensive study on experimental human scurvy by PETERS et al (1948). Not one of the ten patients developed anaemia despite the fact that one had reached the stage where ecchymosis began to appear.

Just prior to this CARTWRIGHT (1947) reviewed the literature on the role of ascorbic acid in haematopoeisis. "The role of ascorbic acid in erythropoeisis is not clear. Although the scorbutic state in both guinea-pigs and human beings is frequently accompanied by anaemic it is questionable whether the anaemia is due specifically to a deficiency of ascorbic acid. Much of the animal experimentation is inconclusive as pure ascorbic acid supplements were not used. Further work in animals is needed. In man it has been both asserted and denied, that synthetic ascorbic acid is effective in relieving the anaemia. It would seem however that there are some scorbutic patients who respond specifically to pure ascorbic acid. The anaemia has been reported as macrocytic, normocytic and microcytic. An induced uncomplicated ascorbic acid deficiency in a human being did not result in anaemia."

The utter confusion caused by these conflicting views on the relation of vitamin C to blood formation in man

is reflected in TOTTERMAN's (1949) and SHAFAR's (1949) review of the position last year.

It is not surprising therefore on consulting various standard text books of general medicine, tropical medicine, haematology and even of the avitaminoses that different accounts are found as to the specificity, severity or even the occurrence of anaemia in adult scurvy.

CHAPTER VI.

THE ANAEMIA OF SCURVY.

THE INCIDENCE, SEVERITY AND SPECIFICITY.

An East African Bantu male was admitted to the medical wards with pain in the limbs, an ulcer on the ankle and a packed cell volume of 14%. Considerable disappointment was felt when the minimal sickling that was produced, could be produced on any blood using that particular oxalate solution. Other causes for this severe anaemia had to be sought. The following day, a megaloblastic bone marrow and a histamine-fast achlorhydria was added to a peripheral smear, that showed marked anisocytosis, poikilocytosis and a mean cell diameter of 8.494 u. On the fourth day, whem liver extract was to be given, it was noticed that the reticulocyte count had shot up to 20%. Then followed rapid improvement clinically and haematologically with the megaloblastic bone marrow reverting to normal. The only factors, that could have been responsible for this dramatic response, were a mixed hospital diet and vitamin C, the latter having been given at the outset for the follicular hyperkeratosis and "spongy" gums. Fasting for the fractional test meal meant that this response, had it been due to the hospital diet, would have been only forty-eight hours after some orally administered deficient factor. It seemed logical, then, to conclude that the intravenously administered ascorbic acid was responsible.

The original doubt that such a severe anaemia could be associated with scurvy, especially when no history

Table 5.

Case No.	Age (Yrs.)	Year	R.B.C. Million/ cu. mm.	HB. G.%	P.C.V. %	Reticulocytes %	W.B.C. per cu. mm.
14.	60	1949	5.3	16	51		8,880
15.	32	1949	4.7	14	42	1	5,900
16.	37	1949	5.3	14	45		6,150
17.	49	1949	2.1	5.2	17		4,280
18.	50	1949	3.6	11.0	31		7,200
19.	30	1949	3.9	12.7	39		3,600
20.	35	1949	2.8	8.5	26		3,920
21.	30	1949	1.7	5.0	14		2,200
22.	25	1948	1.5	5.0	17	+ +	8,360
23.	35	1948	2.8	7.5	24		4,550
24.	52	1948	2.2		25	4.1	8,960
25.	25	1948	0.8	4.0	8	1	2,750
26.	53	1948	1.0		12	18.2	2,900
27.	28	1947	1.8	5.0			4,250
28.	30	1947	3.5	9.4	28		3,950
29.	45	1947	1.6	4.7	14	8.9	6,700
30.	45	1946	2.8				5,600
31.	36	1946	4.3	15			6,650
32.	22	1945	4.0	12			4,500

of blood loss was forthcoming, was not dispelled by consulting the standard text books. Furthermore it was pointed out that no proof existed that this patient's anaemia would have responded completely to ascorbic acid. A full diet, adequate in protein, etc., may have caused the complete regeneration and that the better diagnosis some suggested in this case was "Nutritional Macrocytic Anaemia with Scurvy".

The case notes of all the cases of scurvy admitted to the Hospital during the previous five years were then surveyed. Nineteen cases were available for haematological study. From these (cases 14 to 32 in the total series presented in the first section of this thesis) data were obtained to show that anaemia was of frequent incidence in scurvy and that this anaemia could be very severe. The lowest recorded figure was 8% packed cell volume. It will be seen from Table 5 that only four of these nineteen cases had a red cell count over four million. In five cases (Cases 24, 25, 26, 28 and 29) bone marrow examinations showed a normoblastic hyperplasia although in one of these (Case 26) mention of "numerous megaloblasts" was noted. In two cases (Case 22 and Case 26) normoblasts were seen in the peripheral smear. In the severe cases marked variation in cell size and haemoglobin content was commented upon but otherwise normochromic and normocytic was the usual description. The haematological indices (M.C.V., M.C.H.C. and M.C.H.) reflected this opinion. In four the M.C.V. was over 100, however. In no case could external blood loss have accounted for this anaemia. In

three cases (Cases 24, 25 and 32) there were records of vitamin B complex deficiency as well. Other possible complicating factors were splenomegaly in two (Cases 25 and 31), both East African Natives where malaria is endemic, and tuberculosis in two (Cases 17 and 18) - the former a tuberculous gland proved on biopsy and the latter, a clinical diagnosis of tuberculous epididymitis.

From this survey anaemia appeared to be a far more common association with scurvy and more severe than the text-books would make one believe.

All cases were placed on the hospital diet with vitamin C and usually vitamin B complex added. One case (Case 17) was given iron as well while another (Case 27) received both liver extract and iron. Case 25 with the packed cell volume of 8% was given three pints of blood on admission but two hours later the patient collapsed with a blood pressure of 58/0 mms. Hg. but recovered soon after. The following day his red cell count was lower than on admission. With vitamin therapy this patient's packed cell volume reached 41% on discharge less than a month later. Apart from what has been mentioned in the above three cases no other haematinic drugs were used in this series. All were discharged as cured. Seven cases (Cases 17, 19, 21, 23, 25, 26 and 29) recorded normal blood pictures as positive evidence of this. Four more cases (Cases 20, 22, 24 and 27) had at some time in their progress notes, a markedly improved blood

picture recorded.

Although suggestive evidence that the anaemia was due to vitamin C deficiency might be forthcoming, as yet there was no definite proof that this was the case. In all the above cases a hospital diet had been given. The position with regard to the necessity of ascorbic acid for erythropoeisis was no clearer.

On consulting the current literature, which is reviewed in the previous chapter, the position became even more confusing. Prior to 1935 it was universally believed that vitamin C was necessary for blood formation. After this with pure synthetic ascorbic acid available the position should have become more definite but this has not been the case. This paradox may have resulted from the development of the chemical tests as an assay of vitamin C nutrition. Undoubtedly other factors must have played a part.

The problem is a very difficult one. Firstly scurvy is a deficiency disease and anaemia per se may result from deficient factors in the diet e.g. iron, protein, extrinsic factor, other vitamins etc. Consequently to prove the specificity of one deficiency disorder occurring as a result of another, needs very rigid control. The problem is not made easier by the haemorrhagic tendency in scurvy. Furthermore, it is conceivable that this dietary deficiency may weaken the resistance of the organism to obnoxious factors,

such as infection, which in turn, may distort the true clinical and haematological picture.

There are several avenues of approach to this problem. Firstly there is experimental work on animals; in the case of scurvy, the guinea-pig. Here a diet adequate in all respects but for the deficient factor is given and all the developing features observed. Final proof of the specificity or purity is obtained only if these features promptly improve on supplying that particular deficient factor and no other.

The work on guinea-pigs has shown us that young guinea-pigs are liable to succumb before anaemia develops but that anaemia can readily be produced in older guinea-pigs. However, it is late in onset but it responds to pure synthetic ascorbic acid.

In WOLBACH's words (1937) "the gross and microscopic pathologic changes of human scurvy are so nearly identical that no reasonable doubt can be entertained with regard to applying to the human being the facts ascertained from experimental studies". ZILVA (1944)[KELLIE and ZILVA (1939)] states that from detailed experimental work on the guinea-pig with scurvy, indications have been obtained that a quantitative relationship exists between the guinea-pig and man in several respects.

If one agrees with these statements, one would

expect anaemia to be a late symptom of scurvy and that anaemia would not be so frequent in infants. Such, apparently is the case. Furthermore the course of scurvy in infants is somewhat different from that of adults. Infection, especially, with its effect on the erythron is likely to supervene. Radiological features, so characteristic of infantile scurvy are not seen in adults. Failure to appreciate this difference has probably caused some of the confusion with regard to the necessity of ascorbic acid for erythropoeisis.

The next avenue of approach would be an attempt to produce this feature in the adult human. In those series of experimentally induced vitamin deficiencies the investigator has gone as far as he dare, in an attempt to produce more and more symptoms and signs [GILDER (1950)]. To one investigator such an experiment resulted in his death in 1769 [DRUMMOND and WILBRAHAM (1935)]. The question of wound healing [CRANDON et al (1940)] and certain unpleasant cardiovascular effects [PETERS et al (1948)] made other series cease before reaching the haematoma stage. One volunteer (Crandon) was a blood donor, who lost altogether 6,000 ml. blood over the period of deprivation. The fact that even this did not lead to anaemia is not surprising. During this deprivation period, when this blood loss occurred, scorbutic signs, and then only the asymptomatic skin lesions, were present only in the last six or seven weeks. There were not even gum lesions. Clinically then he would have been classed as a very mild or early case. If we believe what the

guinea-pig tells us, at this stage the erythron is still functioning normally.

An avenue explored by some, has been an approach from the anaemic aspect. In taking a group of anaemic patients these investigators [CROFT and SNORF (1939), LOZNER (1941), LIU et al (1941), SCHULZE and MORGAN (1946) and KEKWICK et al (1947)] basing their conclusions on the response to ascorbic acid in most cases, have attributed the anaemia to vitamin C deficiency on the basis of the chemical tests. These tests have now been shown to be very unreliable.

The only other approach then is from the scorbutic aspect. It would then be necessary to determine the incidence and severity of anaemia amongst the larger series. Furthermore as VILTER et al (1946) show, a mere glance at the oral mucous membranes may be misleading. The gums can be blueish red even with a haemoglobin of 6 or 7 G. The difficulty is enhanced by the fact that the type of patient liable to scurvy, is unlikely to report for medical opinion until the disease is almost incapacitating. Consequently to determine an accurate incidence is not easy.

To prove the specificity of this anaemia is even more difficult. With scurvy as with other deficiency diseases other factors necessary for the organism and/or its erythron may be deficient. The next important consideration is that in the adult, less than 10 mg. per day is all that is necessary to prevent scurvy developing [PETERS et al (1948)]

and 20 mg. per day orally was all that was necessary to cure rapidly a severe case of scurvy [BARNES (1947)]. This has made the position of response to bed rest alone clearer, by diminishing the requirements and allowing any available vitamin C from the body stores to have its effect. This small daily requirement also shows that, if a controlled diet is adopted, this diet must be devoid of vitamin C, as even an intake as low as 5 mg. per day, as in the diet used by RALLI and SHERRY (1941), may be enough to help the body stores supply the body needs. Finally it has been shown that the milder the scurvy and its anaemia, the more adequate are these stores likely to be.

In Chapter V mention was made of work by UNGLEY (1938), RALLI and SHERRY (1941), LOZNER (1941) and McMILLAN and INGLIS (1944). These authors exhibited this phenomenon, but the conclusions drawn, were not in favour of ascorbic acid having any effect on the erythron. This work, of course, was prior to the work of PETERS et al (1948) where exact proof of this small daily requirement was conclusively shown.

From these points emerge then two important pre-requisites for the ultimate proof of specificity:-

1. A diet almost devoid of vitamin C,

2. A control period on this diet alone,
   to determine the effect of bed rest.

To this must be added an eventual response promptly

and completely to pure synthetic ascorbic acid. This would make the excellent work by METTIER et al (1930) invalid, as orange juice contains vitamin B complex and iron as well as vitamin C.

The question of the type of controlled diet is a debatable one and many varieties have been used by several investigators. In the main they are:-

(a) A diet lacking vitamin C but adequate in all other respects. In this way a "gamble" is taken, i.e. one assumes that all features presented by the patient are due to scurvy. Should a response during the control period occur, however, it would be impossible to determine whether it was the result of diminished requirement of vitamin C or the result of some other factor being supplied in the "adequate" diet. On the other hand, if no response or, what would be even more convincing, a deterioration occurred during this control period, followed by an immediate and prompt response subsequently on supplying vitamin C, then it could be assumed that the responding features were due to vitamin C lack.

(b) A diet to the other extreme from (a), i.e. not only lacking in vitamin C but lacking in all known factors that could possibly play a part,

e.g. protein, iron, extrinsic factor and other vitamins. If such a diet is continued for long enough new deficiency syndromes may arise to complicate and confuse the picture further.

(c) A diet which approximates as near as possible, qualitatively and quantitatively as well as in the method of preparation, the diet on which the disease developed. In this way a response during the control period would presumably be due to diminished requirement of vitamin C. A full and complete response on adding only ascorbic acid would undoubtedly prove specificity. On the other hand this diet would allow one to add various haematinic factors e.g. vitamin B_{12}, folic acid, iron etc., and observe whether these factors were, in any way, responsible for the associated anaemia or not. If possible, these factors should be given parenterally to obviate any possible failure of absorption present in the scorbutic state.

The third important consideration in assessing specificity is the response itself. It has never been proved that ascorbic acid, by itself, is specific against any condition other than scurvy [FOX (1940), EDDY and DALLDORF

(1941), SHAFAR (1949)]. The findings of DYKE et al (1942) have been mentioned and reference has been made that scurvy may have co-existed. Conflicting reports of the results of ascorbic acid with desoxycorticosterone acetate (DOCA) on Rheumatoid Arthritis led to an investigation into the effect of ascorbic acid only, in 1000 mg. doses intravenously on several cases, severely anaemic from other causes. (Five of these case histories are briefly noted in Appendix II). No response whatever occurred. VAUGHAN (1934) found no response in other anaemias. TOTTERMAN (1949) further showed that ascorbic acid had no non-specific effect on the erythron by obtaining no response in the anaemia of infection nor in individuals with a normal blood picture. The only change noticed was an increase in the blood ascorbic acid levels in those in whom it was at a low level prior to treatment.

MINOT and CASTLE (1935) showed that in anaemia, a reticulocytosis followed by a rise in the red cells, haemoglobin and, of course, the packed cell volume indicates a response. Furthermore the degree of reticulocytosis is proportional to the severity of the anaemia and the adequacy of the factor supplied. Above an initial level of 3 million red cells per cu. mm. no appreciable reticulocyte rise occurs (VAUGHAN (1934)]. To obviate absorption defects the vitamin should be given parenterally. It has also been shown [MINOT and CASTLE (1935)] that where two deficiencies e.g. Liver and Iron co-exist, the anaemia may respond up to a

point with one factor and not be completely cured until the other factor is supplied. Consequently to prove that the anaemia accompanying scurvy is due to vitamin C lack only, a complete response should occur when only ascorbic acid is added. This fact would be a further criticism against the diet (a) mentioned previously. The presence of iron, protein, etc., in this "adequate diet" may have been responsible for the completeness of the response. This possibility could not be excluded in the conclusion reached by VILTER et al (1946) who used this type of diet.

It would be reasonable to assume, then, that a prompt and complete response as shown by a rise in the packed cell volume with or without a reticulocytosis, following the addition of pure synthetic ascorbic acid only, preferably given in large doses parenterally, to a controlled diet would indicate specificity of the anaemia in scurvy. The best dietary control is a diet on which the disease developed or a diet lacking all factors that may play a part in haematopoeisis providèd, in the latter, that it does not induce other deficiency syndromes which may complicate the picture. A preliminary period of bed rest only is suggested, to determine whether any response to diminished requirement occurs. Finally, it must be stressed, again, that attributing anaemia to vitamin C deficiency, purely on the basis of the chemical tests is unwarranted. The weight of evidence points to a late onset of anaemia long after clinical features become manifest.

AUTHOR	METTIER et al (1930)			VAUGHAN (1934)	DUNLOP et al (1935)	JENNINGS et al (1938)		McMILLAN and INGLIS (1944)					VILTER et al (1946)			BARNES (1947)
No. of Cases	9			I	2	2		25					II			I
	5	2	2			I	I	4	II	4	2	3	I	2	6	3	
Diet Used	Mixed	Boiled milk and flour		Vit C free	As received outside	Mixed	Vit C and iron free	Mixed		Vitamin C free				Vit C free: Low vit B complex			Sterilized milk
RESPONSE FOLLOWING (a) Bedrest only		Nil	Nil & a fall	Fall	Nil in one Slight rise in other	Nil				Slight rise	Nil	Nil	Nil	Slow rise	Nil	Fall	
(b) Iron		Nil & a fall				Nil				Slight rise (cont.)	Nil		Nil				
(c) Liver			Nil			Nil							Nil				
(d) Others						Nil Blood							Nil Thyroid				
VITAMIN C (a) Non-Synthetic	in diet	Orange Juice 575 ml daily	Fresh Liver Full diet					in diet 15 mg									
(b) Synthetic				50 mg oral	60 mg oral	600 mg oral		Nil	500 mg. oral					500 mg oral or parenteral			20 mg oral
Response	Complete	Prompt and Complete		Prompt and Full	Prompt & Complete	Prompt & Full		Prompt		Prompt in 5:2 already normal			Nil (? low normal)	Prompt and Complete			Prompt and Complete
BASIS FOR VITAMIN C DEFICIENCY	Clinical			Clinical	Clinical	Clinical		Clinical						Clinical			Clinical

ISRAELS (I943)	GOTTLIEB (I945)	LOZNER (I94I)				CROFT (I939)	RALLI (I94I)	KEKWICK (I947)		DYKE et al (I942)	UNGLEY (I938)		
3	3	5				6	2	I6		3	3		
		I	I	I	2			9	7		I	I	I
No details given	Mixed	Vitamin C free Vitamin B Complex free				Mixed	Vit C free	Vitamin free Low protein		?Mixed	Vitamin C low (How low not stated)		
		Nil	Rapid rise: Then nil	Slight rise				Nil	Nil		Rise	Slight rise	
		Nil		Nil	Rise								
										Fall			
						In diet					That in diet		
Large doses	700 mg parent-ally	400 mg oral	400 mg oral	400 mg yeast & iron	400 mg oral	I00 mg oral	4000 mg paren.	700 mg orally		I00 mg oral	50 mg oral	600 mg oral	50 mg; then a week later 700 mg oral
Prompt & Full	Prompt & Full	Prompt & Complete	Nil	Rise	Nil	Nil	Prompt	Rise	Nil	Good	Blood already normal	Prompt and Complete	
Clin-ical	Clin-ical	Clinical and Chemical	Clinical Chemical (Post-venesection)		Chem-ical only	Chem-ical only	Clin-ical	Chemical no clinical details		No details given	Clinical		

The results of this detailed analysis of the literature on the anaemia in scurvy reported in the previous chapter are summarised in Table 6. Unfortunately many authors have not given adequate details with regard to those points mentioned above. For reasons already given, reports on infantile scurvy are omitted.

One need not wonder why the question of specificity of ascorbic acid in the anaemia accompanying adult scurvy is a debatable one when one consults Table 6. Only three investigators did not take the "gamble". Only one of these three, DUNLOP and SCARBOROUGH (1935) placed the patient on the diet on which the disease developed. METTIER et al (1930) used boiled milk and flour but this was prior to the synthesis of pure ascorbic acid. BARNES (1947) used sterilized milk but failed to observe any control period. Six investigators used mixed or hospital diets on twenty-two patients and in one other series no details of the diet used were given. These consequently prove nothing. On forty-nine patients eight investigators used vitamin C low or vitamin C free diets. Four of these used diets adequate in other respects [VAUGHAN (1934), UNGLEY (1937), RALLI and SHERRY (1941), McMILLAN and INGLIS (1944)]. In only one of these [VAUGHAN (1934)] is there no doubt that the anaemia was due to vitamin C lack, shown by a deterioration in the blood picture during the control period and a prompt and complete response to ascorbic acid. The remaining four investigators used diets which were low in vitamin B complex in two [LOZNER (1941), VILTER

et al (1946)], free of iron in a third [JENNINGS and GLAZEBROOK (1938)] and free of all vitamins with low protein content in the fourth [KEKWICK et al (1947)]. Three cases in VILTER et al's series (1946) show deterioration over the control period and a prompt and complete response to ascorbic acid. One other case kept on a vitamin C free "adequate" diet by RALLI and SHERRY (1941) for fifty-two days was not included in the table as no haematological details were given apart from the initial findings and a remark that the blood picture had not deteriorated over this period.

Using these criteria it is shown that in very few cases indeed has the specificity of ascorbic acid in the anaemia accompanying scurvy been established without doubt. Combined deficiencies in many cases cannot be ruled out. On the other hand those who hold the view that vitamin C is not specific in the uncomplicated anaemia of scurvy, hold it on very flimsy evidence.

In an attempt to settle this question the following plan was adopted for the rest of the cases in this series.

1. Care was taken that no unprescribed ascorbic acid reached the patient.

2. Each patient was placed on the diet on which he developed the disease. This was facilitated by the fact that all had eaten a similar diet, the staple diet of the Bantu here. This was mealie meal porridge without

Table 7.

Case No.	Age (Yrs.)	Year	R.B.C. Million/ cu. mm.	HB. G.%	P.C.V. %	Reticulo-cytes %	W.B.C. per cu.mm.
1.	30	1949	1.5	4.6	14	2	3,400
2.	37	1949	3.9	14	37	1.2	7,040
3.	40	1949	2.0	6.0	20	1	8,600
4.	49	1949	2.2	7.0	23	8	6,500
5.	25	1950	2.7	8.2	22	1.3	12,200
6.	28	1950	1.8	5.4	15	4.1	3,900
7.	50	1950	6.3	18	59	0.6	7,150
8.	35	1950	2.7	7.3	23	7.4	7,200
9.	24	1950	2.8	7.3	23	1.8	8,150
10.	60	1950	2.8	8.6	26	3.3	7,600
11.	27	1950	4.3	13.6	42	1.4	6,700
12.	55	1950	2.9	8.8	27	1.6	6,000
13.	59	1950	1.5	4.6	15	7	6,200

milk or sugar, alternating with "stamped" mealies, bread without butter or jam, black tea, coffee or water. This was given three times a day.

3. Detailed haematological investigations on admission are reflected in Table 7. It will be seen that of these, all consecutive admissions where no selection prior to admission was possible, only two are not anaemic. In one (Case 7) the scorbutic manifestations were mild and in the other (Case 11) the duration of the history was just under three weeks. Three cases had a packed cell volume of 15% or less, and but for Case 2 the remainder varied between 20% and 27%. It is fair to state, however, that this must be considered as the incidence and severity of anaemia in severe scurvy despite the fact that they are consecutive admissions. The patients in this series would report for medical attention only when the disease becomes incapacitating. It would be impossible, due to the type of patient and often language difficulty, to assess accurately whether or not the duration of the disease or dietary deficiency influenced the presence or severity of the associated anaemia of these cases of adult scurvy. It was an impression, however, that the duration of the disease did have this influence.

4. Synthetic ascorbic acid given parenterally in 1000 mg.

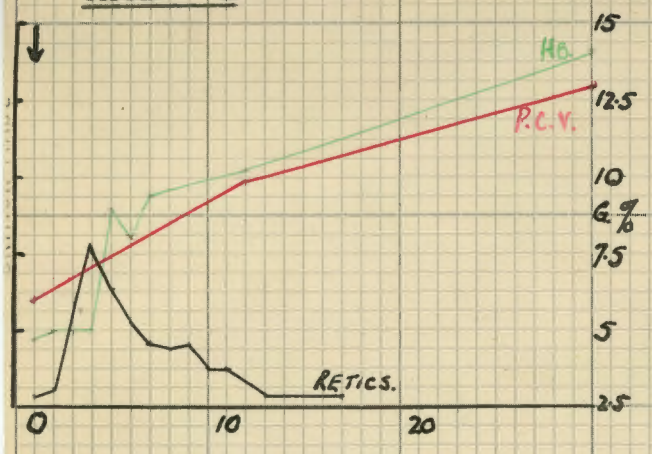
daily doses was used to assess specificity whilst still on this control diet. This diet was continued until the packed cell volume was in the region of 40%. In one case (Case 8) 300 mg. ascorbic acid was given by mouth divided into 100 mg. doses daily over three days. This was given to see whether iron, given intravenously, would produce a response after ascorbic acid was given. In Cases 6 and 9 it produced no response prior to ascorbic acid therapy.

Variable control periods were used. In Cases 1, 4 and 12 the vitamin was given on admission. Case 1 was the case described at the beginning of this Chapter and, as stated, consumed a mixed hospital diet as well. The shortest control period was in Cases 2 and 3, being only two days. The other cases had control periods varying from seven to thirty days.

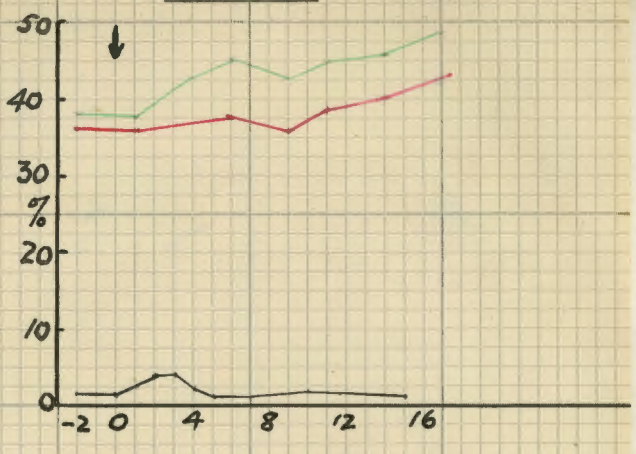
During this time Cases 6, 8 and 9 were given vitamin B_{12} in 30 u G. doses daily for four days, by injection, folic acid 15 mgm. daily for five days by mouth, iron in 400 to 500 mg. doses divided over five days intravenously and vitamin B complex as "Plebex" or "Bejectal" in 2 ml. doses parenterally. Sufficient time was allowed to elapse between each of the above, to observe any haematological response.

One case, confined to bed during the control period

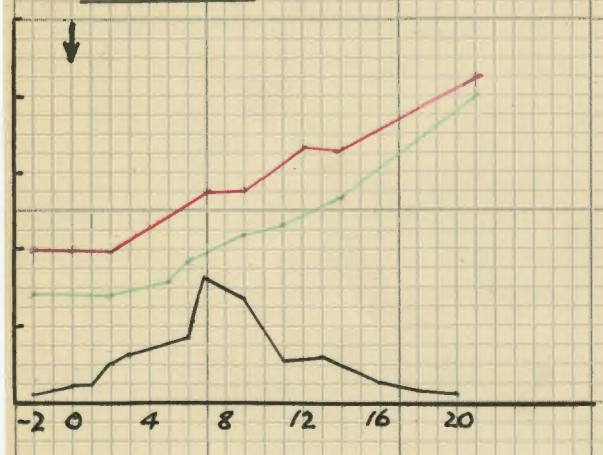
CASE 1



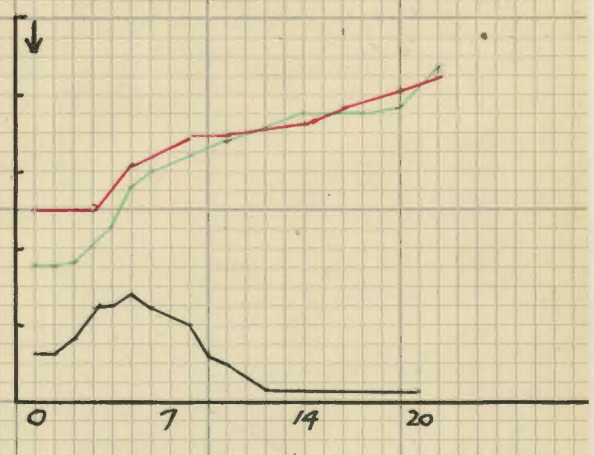
CASE 2



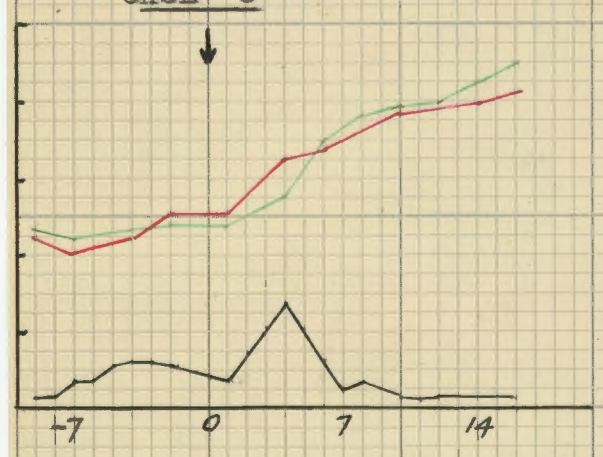
CASE 3



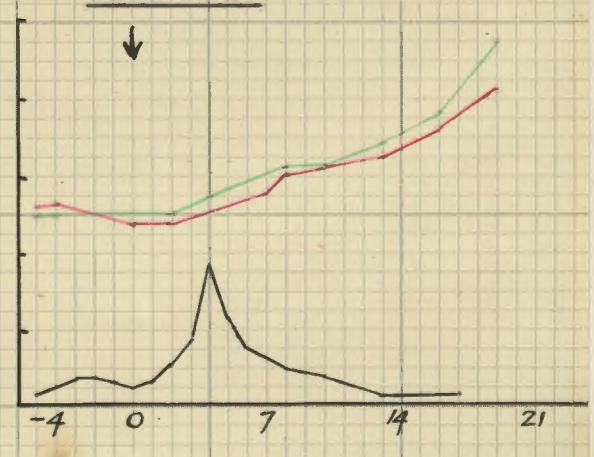
CASE 4



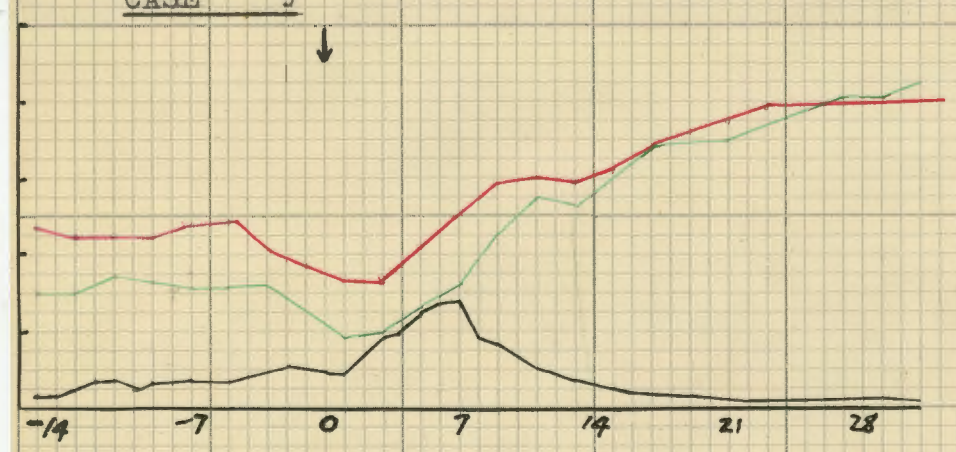
CASE 5



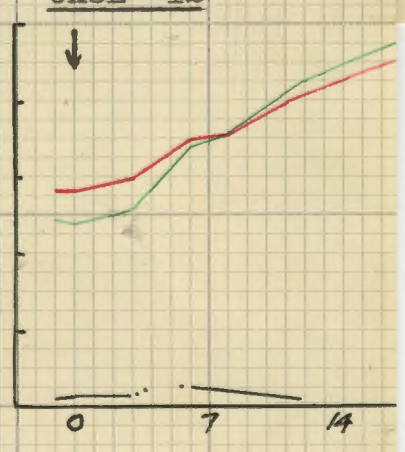
CASE 10



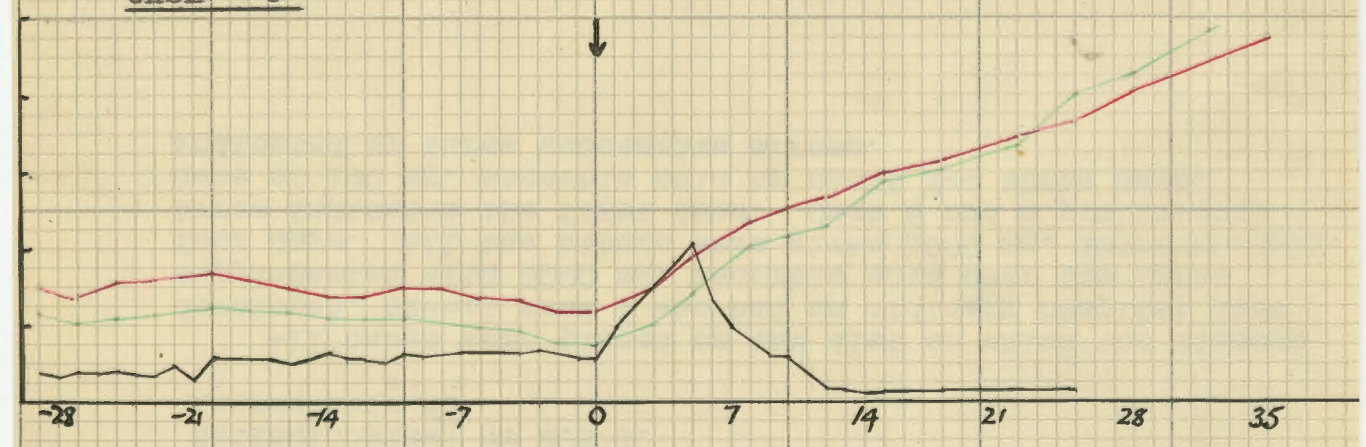
CASE 9



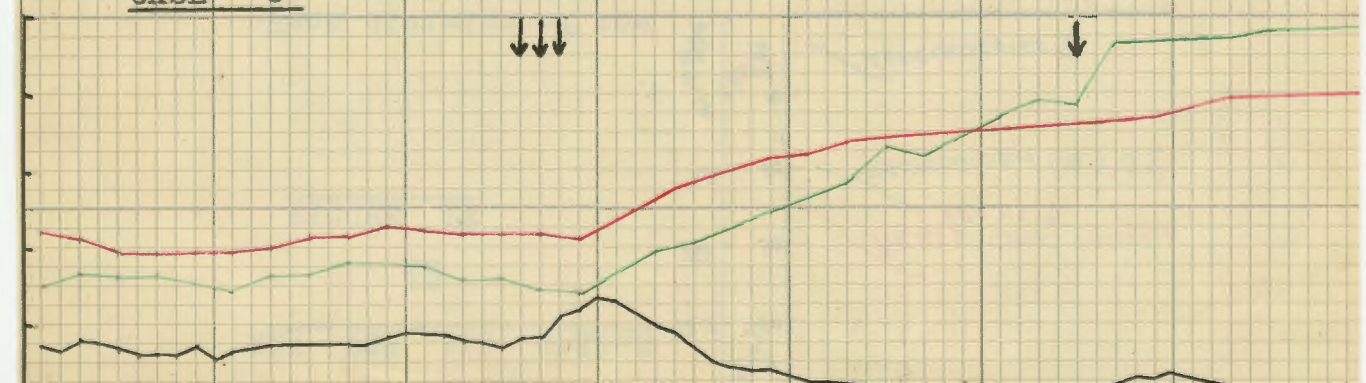
CASE 12

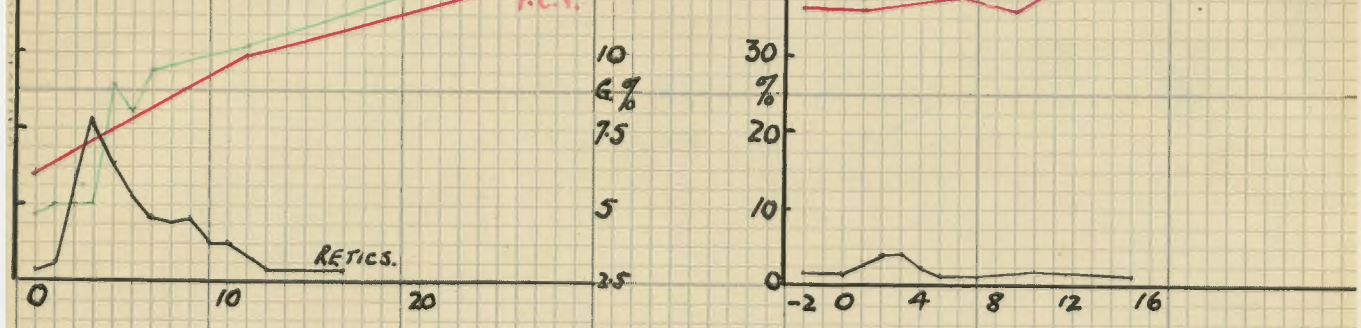


CASE 6

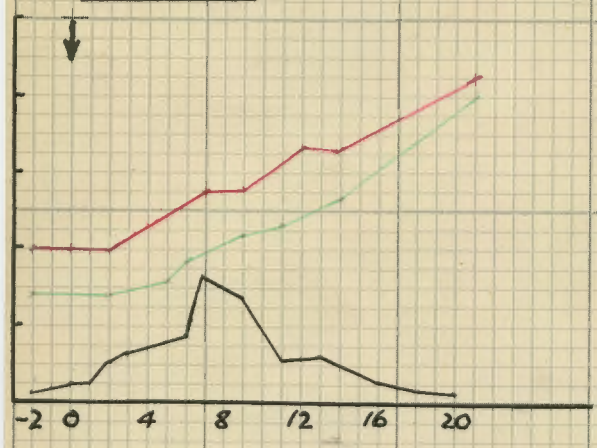


CASE 8





CASE 3



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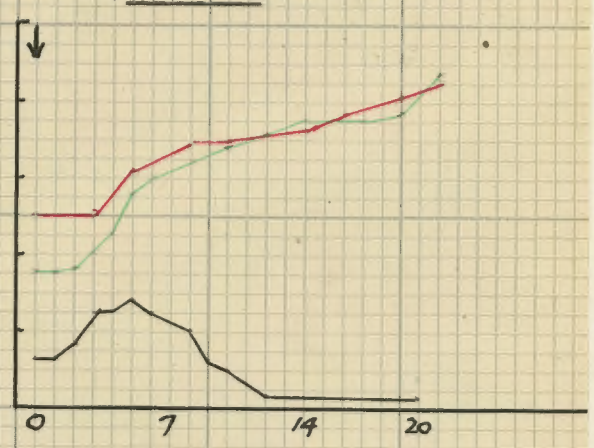

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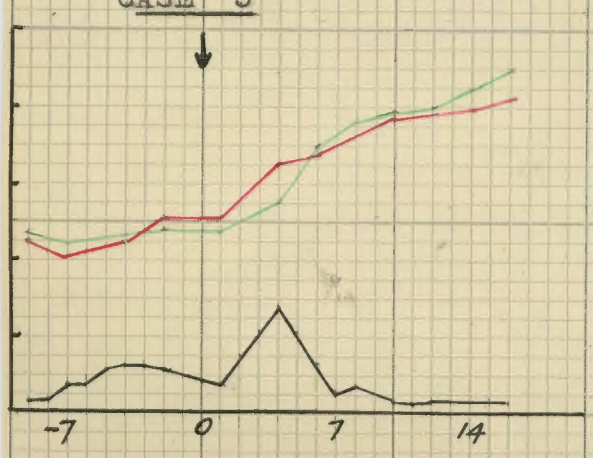
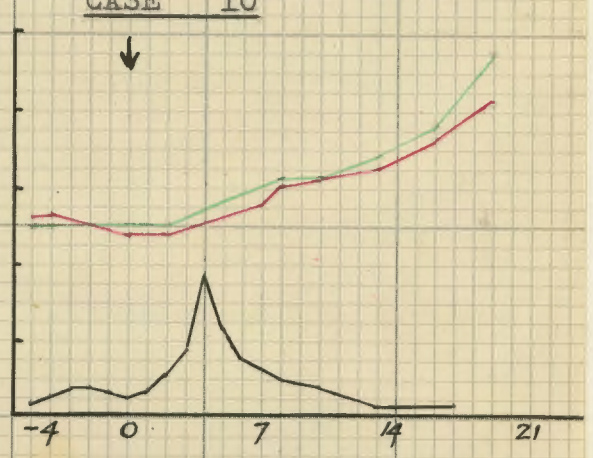
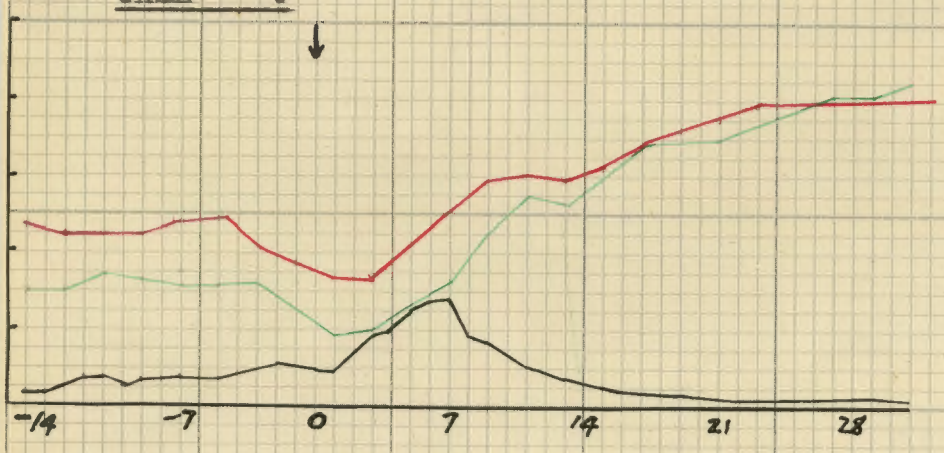


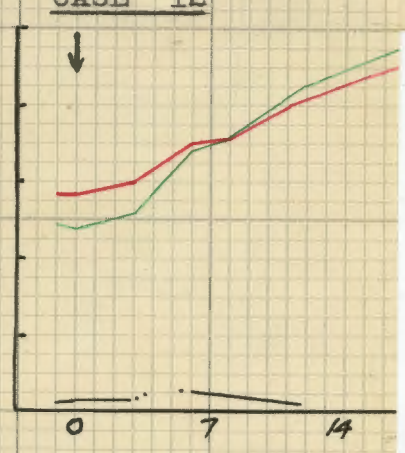
CASE 10



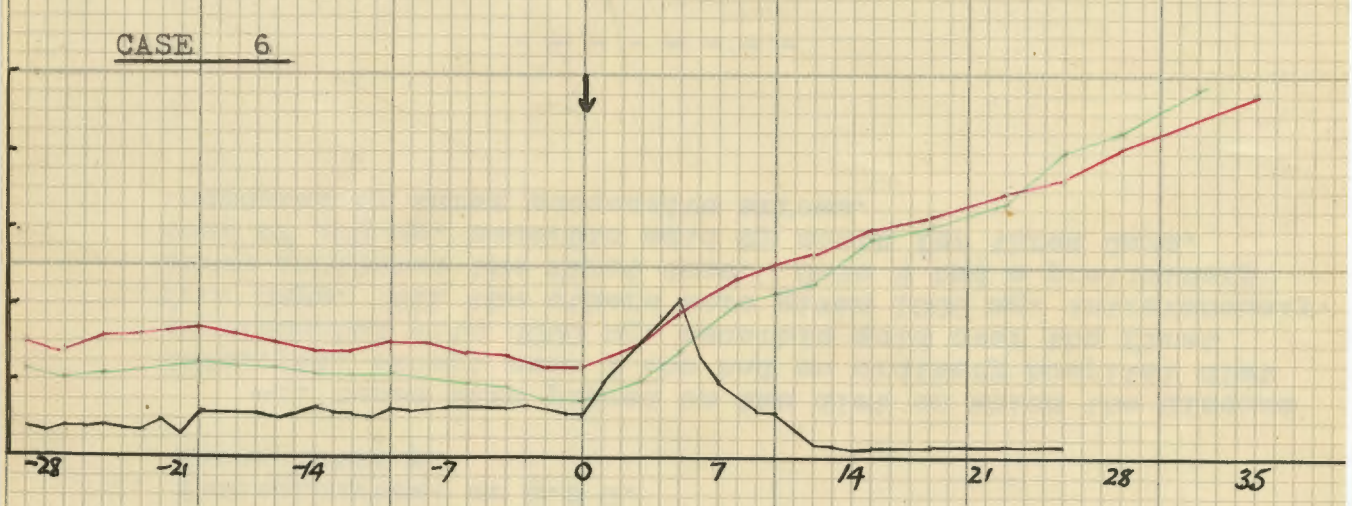
CASE 9



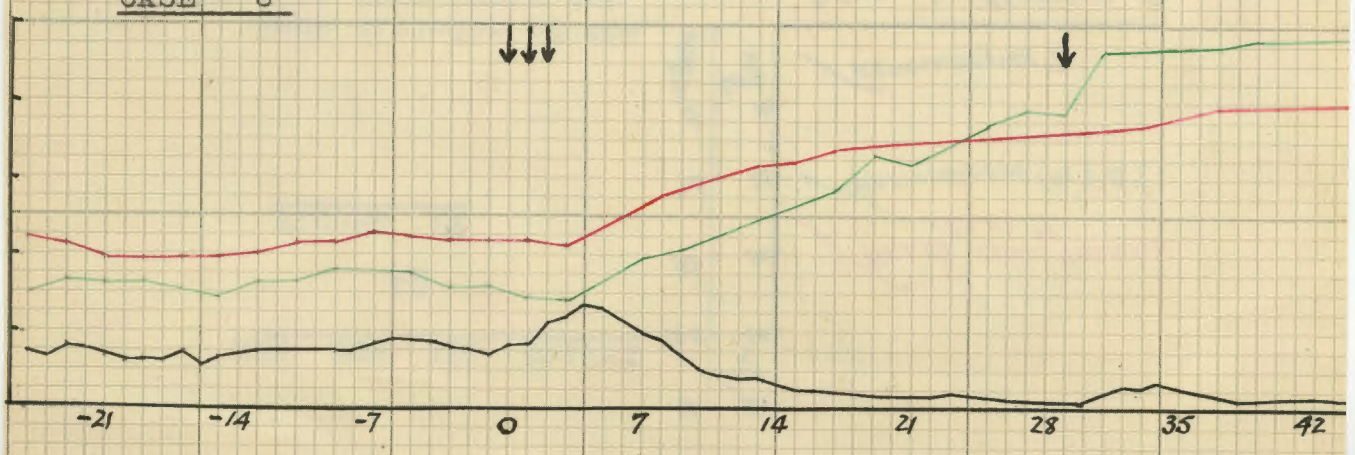
CASE 12



CASE 6



CASE 8

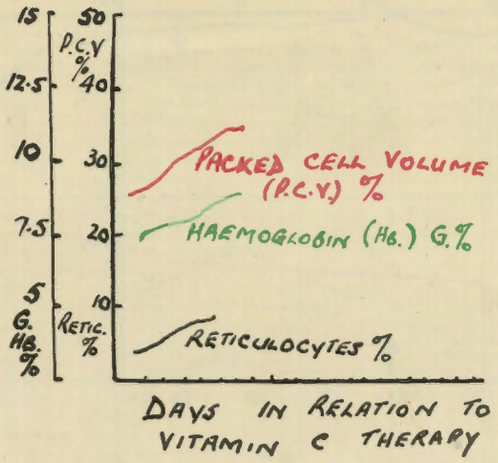


DAYS IN RELATION TO VITAMIN C THERAPY

KEY FOR ALL GRAPHS

IN

FIGURE 1



Each division of the abscissa represents one day. Four divisions of the ordinate represent either 10 % (P.C.V. or Reticulocytes) or 2.5 G. Haemoglobin %.

All cases were placed on the diet on which the disease developed, except case 1 who had an ordinary hospital diet supplemented with 3000 mg. ascorbic acid over the first four days. All the others were given 1000 mg. intravenously. Arrow indicates the first dose. Case 8 had a preliminary dose of 100 mg. ascorbic acid by mouth for three days, indicated by three successive arrows.

- - - - - - - -

(Case 5) was allowed up once ascorbic acid therapy was given to see whether this would interfere with the rapidity of the response. Another case (Case 12) was by force of circumstance treated as an outpatient, attending bi-weekly for haematological investigation and ascorbic acid injections. An independent observer was asked to ensure that no alteration in the original diet had occurred.

The results of this investigation are graphically represented in Figure 17, in the ten anaemia cases followed up. It will be seen that all except two cases showed a prompt reticulocytosis reaching the peak between the fourth and sixth days after ascorbic acid was given. This was followed in each case by a rapid and complete haematological recovery. The two cases (Cases 2 and 12) showing no significant reticulocytosis were the least anaemic of the series. The response in these was judged on the rise in packed cell volume.

The packed cell volume in one case only, (Case 5), rose during the control period although two others (Cases 6 and 9) showed a preliminary rise over the first ten days and then began to fall. It was felt that this rise could have been due to diminished requirement of vitamin C by bed rest. In Case 5, however, an epistaxis, apparently not severe, brought the patient to hospital. No bleeding occurred while the patient was hospitalized consequently this response may have been regeneration from blood loss. In comparing

the ten day periods before and after treatment there can be no doubt as to the response to ascorbic acid, i.e. 4% rise in packed cell volume as against the 12½% rise after treatment.

If one excludes Case 1 as a mixed hospital diet was given, there can now be no doubt whatever that ascorbic acid is necessary for erythropoeisis. It is fair to assume that the anaemia accompanying scurvy in this series is due solely to a deficiency of vitamin C. A prompt and complete response occurred without any other factor such as adequate amino acid intake, iron, extrinsic factor or other vitamin, being necessary before, during or after ascorbic acid administration, whether they were deficient, at the same time, in the diet or not.

Finally, it is suggested that the previous confusion with regard to the role played by ascorbic acid in haematopoeisis is due to:-

1. Failure to observe the fact that ascorbic acid has no proven effect against any condition other than vitamin C lack.

2. Failure to observe the fact that anaemia due to vitamin C lack is usually due to chronic vitamin C lack in the form of clinically obvious scurvy and that all chemical tests, (with the possible exception of the white cell-platelet layer) are

unreliable in the diagnosis of vitamin C lack.

3. Failure to use pure synthetic ascorbic acid.

4. Failure to use a suitable control diet.

5. Failure to adopt a period of control either to see if diminished requirement of the vitamin by bed rest or spontaneous regeneration from blood loss occurs, and/or to eradicate possible influences exerted by other deficiencies.

C H A P T E R VII.

THE NATURE OF THE ANAEMIA IN SCURVY.

INTRODUCTION.

Although as a result of the previous chapter it may be acceptable that anaemia is a specific feature of vitamin C lack in Scurvy, the anaemia need not necessarily be due to a pure lack of this substance in the body. By virtue of its physiological properties, vitamin C, with its possible participation in enzyme reactions [SHAFAR (1949)], may be necessary before some other factor can be utilised in erythropoeisis, e.g. the presence of vitamin C may be necessary before iron can be absorbed, etc.

Furthermore scurvy has a bleeding tendency consequently the anaemia may arise purely on a post haemorrhagic basis.

Finally, vitamin C may be necessary to prevent premature red blood cell destruction, i.e. haemolysis.

An attempt has been made in this thesis to explain the most likely manner in which vitamin C affects the erythron. It must be appreciated from the beginning that this explanation can only be presumptive on the methods available. Detailed pigment and red cell studies with radio active isotopes are not yet possible in this country.

THE MORPHOLOGY OF THE ANAEMIA OF ADULT SCURVY.

A. THE RED BLOOD CELL.

1. The size, shape and Haemoglobin content of the Red cell.

In Chapter V mention was made that most authors feel that the anaemia of scurvy is normocytic and normochromic. McMILLAN and INGLIS (1944) describe six cases of microcytic hypochromic, fourteen as simple microcytic and two macrocytic in their forty cases. A slight hypochromic anaemia is mentioned by SHULTZER (1936) whereas three of VILTER et al's (1946) eleven cases had mean corpuscular haemoglobin concentrations of 29% and 30%. Their lowest mean corpuscular volume was 82 cu. u. but four patients had a M.C.V. above 96 cu. u.

Using the colour index GOTTLIEB (1945) reports three cases of high colour index anaemia due to scurvy, comparing them with the high colour index of NISENSON and COHEN's (1937) case.

Unfortunately most authors have assessed the size of the red cell on the mean corpuscular haemoglobin and the colour index which are only of limited value [WHITBY and BRITTON (1942), WINTROBE (1946)]. VILTER et al (1946) approach greater accuracy by using the haematocrit. JENNINGS and GLAZEBROOK (1938) used a Price Jones curve to confirm the presence of macrocytosis in one of their two cases. The

only other mean cell diameter estimation recorded was that of VAUGHAN's (1934) case. The reading was 7.05 u with a mean corpuscular volume of 86.4 cu. u.

JENNINGS and GLAZEBROOK (1938) attempt to explain the difference in red cell size as due to the severity of vitamin C lack. The more severe the lack, the further back would maturation be arrested with its consequent larger cell appearing in the peripheral smear.

The comments on the peripheral smears in Cases 14 to 32 in this series were usually normochromic and normocytic. The haematocrit indices (M.C.V., M.C.H.C. and M.C.H.) reflected this opinion. Four cases had an M.C.V. over 100 cu. u whilst the lowest M.C.V. recorded was 81 cu. u. The lowest M.C.H.C. was 31%.

Table 8 illustrates the volume (M.C.V.), diameter (M.C.D.), thickness (M.C.A.T.), haemoglobin content (M.C.H.C.) and the weight of the haemoglobin (M.C.H.) of the average red blood cell of the personal series (Cases 1 to 13). The degree of "flattening" is indicated by the Diameter : Thickness (D : T) ratio, and the packed cell volume reflects the severity of the anaemia in each case. In Cases 2, 3 and 5 no Price Jones curves were performed. Case 7 was not anaemic and for this reason is not included in the table.

From this table it appears that most cases are normocytic and normochromic, having a mean diameter and

Table 8.

Case No.	P.C.V. %	M.C.V. cu. u.	M.C.D. u	M.C.A.T. u	D : T Ratio	M.C.H.C. %	M.C.H. µµ
1.	14	94	8.50	1.76	4.7:1	33	30.9
2.	37	94.8				37.9	35.9
3.	20	101.2				30	30.3
4.	23	106	7.97	2.12	3.8:1	30.2	31.8
5.	22	81.2				36.8	30
6.	15	82	6.92	2.18	3.2:1	36	30
8.	22.5	84	7.18	2.07	3.5:1	32.4	27
9.	23	81.4	7.31	1.94	3.8:1	32	26
10.	26	94.2	7.59	2.09	3.7:1	33	31
11.	41.5	96.5	7.34	2.28	3.2:1	32.8	31.6
12.	27	93.1	7.36	2.19	3.4:1	32.6	30
13.	15	100	7.70	2.15	3.6:1	30.6	30.6

thickness within normal limits. Macrocytic figures are seen in three cases (Cases 1, 4 and 13) and a raised M.C.V. in a fourth (Case 3) which has no mean cell diameter for confirmation. The lowest M.C.H.C. is 30% and in no case was microcytosis observed.

Apart from Case 6 these results together with those noted in the rest of the Total Series (Cases 14 to 32) would give the impression that the more severe the anaemia, the more likely is macrocytosis to be found. An analogous opinion is held by JENNINGS and GLAZEBROOK (1938) who relate macrocytosis to the severity of vitamin C depletion.

Although liver dysfunction was present in variable degrees in the cases studied it is unlikely to have contributed to the degree of macrocytosis as the D:T Ratio was within normal limits in all cases but one. Target cells, furthermore, were rarely seen in the peripheral smear.

Finally, no constant relation between the degree of macrocytosis and the degree of reticulocytosis was present.

2. Variations in the Size, Shape and Haemoglobin content of the Red Blood Cell.

It has been shown that anisocytosis and poikilocytosis become quite marked as the anaemia of scurvy progresses in the guinea-pig [SIGAL (1939)].

McMILLAN and INGLIS (1944) noted that erythrocytes in adult scurvy were mainly normal in size and shape with anisocytosis noted in about 20% of smears, being much commoner than poikilocytosis. VILTER et al (1946) also noted that the red cells varied only slightly in size and shape . DUNLOP and SCARBOROUGH (1935) record marked anisocytosis,

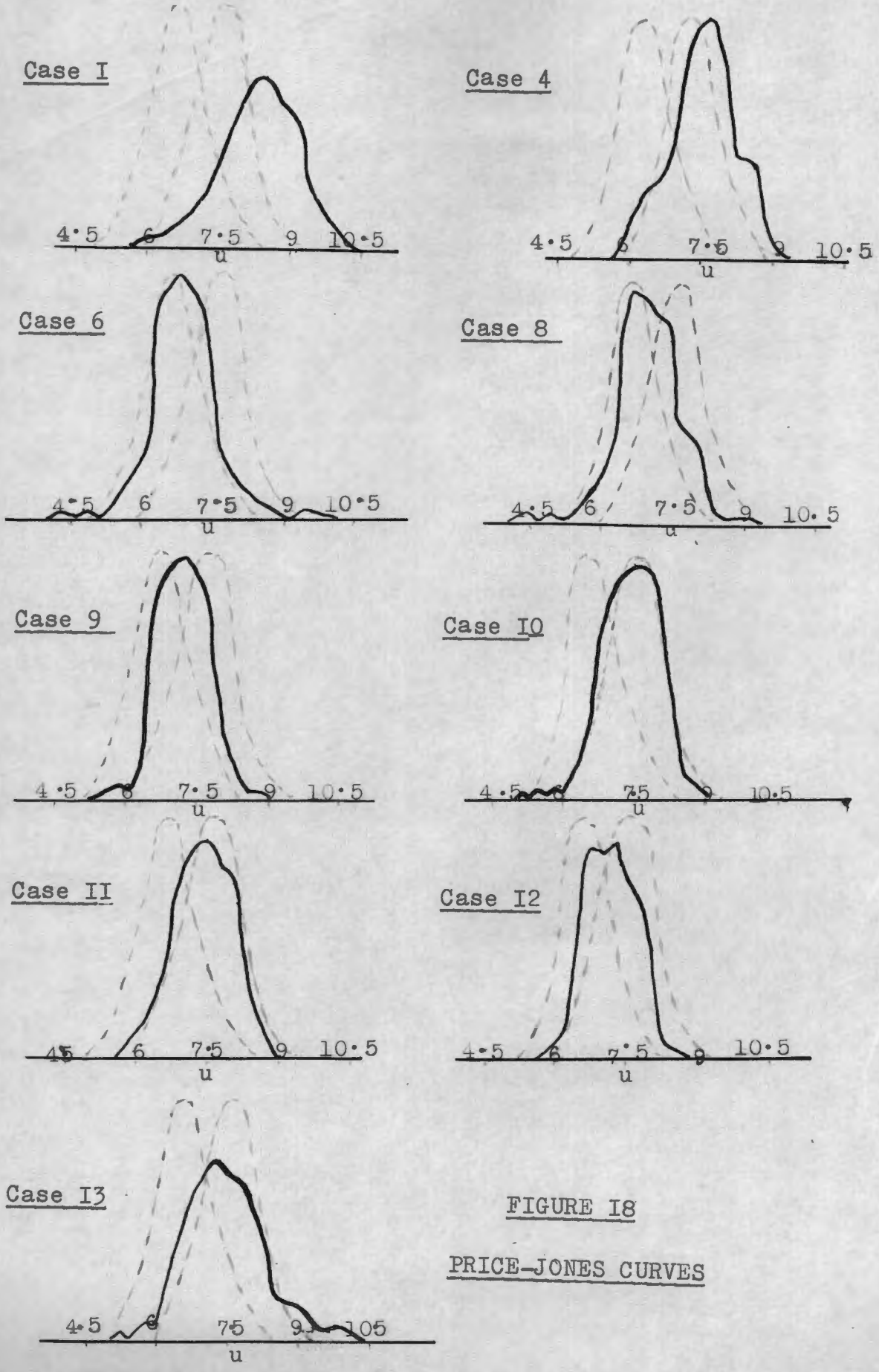


FIGURE I8

PRICE-JONES CURVES

slight poikilocytosis, a number of microcytes and megalocytes in their severe case and a normal picture in the less severe case. Polychromasia, anisocytosis, anisochromia with other features, caused JENNINGS and GLAZEBROOK (1938) to diagnose pernicious anaemia in their severe case. Similar peripheral smears have been reported in severely anaemic cases of scurvy [YOUNG (1938), NISENSON and COHEN (1937), HATHERLEY (1947)].

In Cases 14 to 32 in the total series it was noticed that the severer the anaemia the more marked was the variation in size, shape and haemoglobin content.

In Cases 1 to 13 (i.e. the Personal series) anisocytosis was seen more commonly in the severer anaemias. Poikilocytosis was less frequently seen and was only a feature of the very severe cases. In these cases too, gross variation in the haemoglobin content of the cells occurred. Often large cells were well filled with haemoglobin whilst smaller cells were poorly filled and vice versa. Gross hypochromia was not seen. "Anisochromia" [JENNINGS and GLAZEBROOK (1938)] was the term used to describe the variation in haemoglobin content. "Target" cells were rarely seen.

In the less severe cases gross alterations in the size, shape and haemoglobin content of the red blood cells was not a feature.

The Price Jones curves of nine of the thirteen cases (Figure 18) depict the anisocytosis found, whilst Table 9 illustrates this by the raised figures for the standard

deviation and coefficient of variation. This table also confirms the impression gained from the peripheral smear.

Table 9.

Case No.	P.C.V. %	M.C.D. u	Standard Deviation (u)	Coefficient of Variation (%)	% Macrocytosis
1.	14	8.50	0.869	10.2	56.6
4.	23	7.97	0.689	8.65	22.2
6.	15	6.92	0.695	10.1	
8.	22.5	7.18	0.701	9.75	
9.	23	7.31	0.518	7.1	
10.	26	7.59	0.620	8.19	
11.	41.5	7.34	0.485	6.6	
12.	27	7.36	0.478	6.5	
13.	15	7.70	0.890	11.6	9.2

The more severe the anaemia, the more raised are the figures for the standard deviation and coefficient of variation. Normal figures (i.e. between 0.4 and 0.5 u for the standard deviation and between 5.3 and 7.3% for the coefficient of variation) are seen in the least anaemic cases (Cases 11 and 12). It would appear, then, that anisocytosis becomes more noticeable as the anaemia of scurvy progresses.

3. The Reticulocytes.

In the guinea-pig METTIER and CHEW (1932) have shown that with progression of the anaemia more and more

reticulocytes and nucleated red cells are seen in the peripheral smear.

In man a high reticulocyte count without treatment has been recorded by many [LOZNER (1941), NISENSON and COHEN (1937), JENNINGS and GLAZEBROOK (1938)] in the severe stage of untreated scurvy. VILTER et al (1945)(1946) attribute this to haemolysis, JANET VAUGHAN (1934) to the stimulating effect that repeated haemorrhages have on the bone marrow, whilst DUNLOP and SCARBOROUGH (1935) with METTIER and CHEW (1932) attribute it to failure of the bone marrow to maintain mature cells in the peripheral circulation.

McMILLAN and INGLIS (1944) found no increased number of reticulocytes in their series, but no anaemia was classed as severe.

Five reticulocyte counts were available for study in cases 14 to 32. Four of these cases were severely anaemic and three of these had raised reticulocyte counts. The fourth case was the most severe case of the whole series with a packed cell volume of 8%. His reticulocyte count was less than 1%. One of the cases with a raised reticulocyte count had normoblasts in the peripheral smear. In one other case, with severe anaemia and normoblasts in the smear, the reticulocytes were recorded as + + but no count was available.

Two of these cases were followed up and both showed a reticulocytosis following therapy with ascorbic acid.

In the personal series (Cases 1 to 13) raised reticulocyte levels were seen in the initial haematological investigation in five (Cases 4, 6, 8, 10 and 13)(See Table 10). The reticulocytosis did not appear to have a constant relation to the degree of anaemia.

Table 10.

Case No.	P.C.V. %	Reticulocytes %
1.	14	1.2
2.	37	2.0
3.	20	1.0
4.	23	8.0
5.	22	1.3
6.	15	4.1
7.	58	0.3
8.	22	7.4
9.	23	1.6
10.	26	3.3
11.	41.5	1.4
12.	27	1.6
13.	15	7.0

In all except three cases there was a rise in the reticulocyte count soon after admission whether treatment with ascorbic acid was given or not. With ascorbic acid medication a prompt further rise in reticulocytes occurred with the peak between the fourth and sixth days. This was graphically shown in Figure 17 in the last chapter. The three exceptions (Cases 2, 7 and 12) had the highest packed cell volumes of the series. In three cases (Cases 1, 5 and 13) normoblasts were noticed in the peripheral smear.

B. THE WHITE BLOOD CELLS IN SCURVY.

Most authors seem to agree that the white cell count is either low normal or low in scurvy. METTIER et al (1930) and RALLI and SHERRY (1941) state that the white count usually lies between 4,000 and 6,000 cells per cu. mm. In twelve patients of the nineteen in VILTER et al's (1946) series the white cell count was below 6,000 per cu. mm., one case being 2,500 cells per cu. mm. Although not invariably the case, the lower counts were associated with the severer anaemias. JENNINGS and GLAZEBROOK (1938) noted in their severe case, a white cell count of 3,200 cells per cu. mm. A similar level was noted in NISENSON and COHEN's case (1937).

In Crandon's experiment [CRANDON et al (1940)] although no anaemia developed the white cell count remained between 3,200 and 5,000 cells per cu. mm. and rose to 9,000 after vitamin C only was added to the deficient diet.

The packed cell volumes, total white cell counts and the percentage of Neutrophils (N), Lymphocytes (L), Monocytes (M) and Eosinophils (E) in the differential white cell counts of the total series of 32 cases are compared in Table 11.

The lowest counts were associated with severe anaemia but otherwise no consistent relationship existed, e.g. in three of the nine cases in the series with packed cell volumes below 20% the white cell count was above 5,000 cells per cu. mm.

In those cases followed up after treatment with vitamin C, where the white cell count was normal, no change was noted, whereas in those with a low count this did not return to normal until vitamin C was given.

Table 11.

Case No.	P.C.V. %	Total White Cells per cu.mm.	N %	L %	M %	E %	Case No.	P.C.V. %	Total White Cells per cu.mm.	N %	L %	M %	E %
1.	14	3400	59	33	5	2	17.	17	4280	41	44	7	7
2.	37	7040	48	50	2	0	18.	31	7200	77	21	0	2
3.	20	8600	55	45	0	0	19.	39	3600				
4.	23	6500	65	20	5	0	20.	26	3920	34	56	3	7
5.	22	12200	69	22	6	3	21.	14	2200	53	36	9	2
6.	15	3900	45	49	4	2	22.	17	8360	79	20	1	0
7.	58	6500	49	37	11	0	23.	24	4550	61	26	13	0
8.	22	7200	65	32	3	0	24.	25	8960	64	36	0	0
9.	23	8150	57	38	3	2	25.	8	2750	39	61	0	0
10.	26	7600	46	44	10	0	26.	12	2900				
11.	42	6700	45	45	2	7	27.	1.8*	4250	58	42	0	0
12.	27	6000	61	30	8	1	28.	28	3950	54	38	8	0
13.	15	6200	62	32	5	1	29.	14	6700	60	35	4	1
14.	51	8880	46	50	3	1	30.	2.8*	5600	48	43	3	6
15.	42	5900	68	28	3	1	31.	4.3*	6650	58	35	4	3
16.	45	6150	67	27	4	1	32.	4.0*	4500	55	32	6	7

Cases 1, 16 and 17 had 1% Basophils.

* Red cell count in millions per cu. mm.

One case in the whole series had a white cell count above 10,000 (Case 5). This case had epistaxis and

the white cell count returned to normal after six days of rest in bed on the control diet only. His initial reticulocyte count was 1.3%. There was a slight reticulocyte response during this period of bed rest and in the previous chapter it was concluded that it would be difficult to differentiate diminished metabolic requirement of vitamin C from cessation of the haemorrhage as the cause of this rise. Possibly the slight leucocytosis suggests the latter, but the patient maintained that he had not lost much blood.

The differential white cell count was in most cases normal. Occasionally a relative lymphocytosis was evident.

C. THE PLATELETS IN SCURVY.

Mention has already been made that a normal number of platelets occur in scurvy.

SUMMARY OF THE MORPHOLOGY OF THE ANAEMIA OF SCURVY.

1. The majority of cases of anaemia of scurvy appear to have a normocytic normochromic picture. It appears that as the anaemia progresses the mean cell diameter becomes larger.

2. The red cells appear to become more variable in size, and haemoglobin content but less markedly so in shape the more severe the anaemia.

3. The more severe the anaemia the more noticeable are reticulocytes and occasionally nucleated red cells in the peripheral circulation. If this is not seen initially it becomes manifest soon after bed rest.

4. A marked leucopaenia may develop when the anaemia is severe.

5. The close morphological relationship to pernicious anaemia has often led to an erroneous diagnosis.

THE BONE MARROW IN SCURVY.

WOLBACH (1937) states that in long standing vitamin C lack in guinea-pigs large regions of the bone marrow become devoid of blood forming cells and the seat of a deposit of homogeneous Amyloid-like material.

METTIER and CHEW (1932) studied the bone marrow extensively in the guinea-pig. Some guinea-pigs were allowed to die with scurvy and three, treated with orange juice, were killed at the peak of the reticulocytosis.

Macroscopically the medullary cavity in the long bones of the untreated guinea-pigs was filled with a uniform greyish-red soft tissue. There was a markedly increased cellularity over normal, with almost complete disappearance of fatty tissue which was replaced largely by cells of the erythropoeitic series. Nucleated red cells, mainly of the normoblastic variety, appeared in large numbers. In spite of this there was but little evidence of active cellular maturation. "Adult erythrocytes occurred in small numbers and only an occasional mitotic figure was noted. The development of the cells seemed to be at a standstill."

The guinea-pigs, sacrificed during the reticulocyte response, showed an increased number of cells containing mitotic figures and a larger number of adult erythrocytes in their bone marrows. "Thus there was distinct evidence of active red blood cell maturation in those treated with vitamin C.

This substance is evidently required in order that a progressive development into mature cells can take place."

In discussing these changes and comparing them with their observation that an increasing number of reticulocytes and immature cells appeared in the peripheral circulation and bone marrow as the disease progressed, they concluded: "This suggests that retarded erythropoeisis may be fundamental, i.e. comparable to a delay or retardation in maturation of the red blood cell. Such a hypothesis is substantiated by the reticulocyte response induced following the ingestion of a diet containing vitamin C by the animal with scurvy; the return of the bone marrow to a normal state of activity and the appearance of normal numbers and kinds of red blood cells in the peripheral circulation."

HARRIS (1928) reported the marrow findings in one post-mortem study of "scurvy-rickets", as areas of gelatinous marrow devoid of blood forming cells with failure of normal erythropoeisis.

PARSONS et al (1933)(1935) felt that vitamin C by its power of reversible oxidation, functioning as an oxygen carrier played an important part in the processes of tissue respiration and metabolism. Its action on the bone marrow was throughout the whole range of maturation from endothelial cell to adult erythrocyte and was not restricted to the normoblast stage as suggested by WITTS (1932).

They felt that the anaemia of scurvy resulted from a general slowing down of the whole process of erythropoeisis which may be so marked that marrow degeneration and aplasia resulted. He explained the megalocytic anaemia they saw as due to disproportional slowing up of red cell development at an early stage. They compared this state with the part played by thyroid.

Furthermore Parsons felt that increased mitosis indicated merely increased production rather than maturation.

Unfortunately they did not support these statements with any descriptions of marrow changes of their own cases as ISRAELS (1943) points out.

There appears to have been no work on the bone marrow findings in infantile scurvy in recent years.

In adults, METTIER, MINOT and TOWNSEND (1930) described moderate cellular hyperplasia with scattered small groups of nucleated red blood cells, with no apparent mitosis. At the peak of the reticulocyte crises, quantitatively more nucleated red blood cells were seen and a few mitotic figures were present in each field of the microscope.

In 1938 Jennings and Glazebrook described normoblastic hyperplasia with an increased number of early red cell precursors. No follow up unfortunately was done. They felt that the marrow was compatible with pernicious anaemia and

and noted achlorhydria even after histamine in this case. They felt that due to the long duration of vitamin C lack maturation arrest occurred at a very early stage.

ISRAELS (1943) described diminished cellularity and failure of erythropoeisis in two cases of scurvy with mild anaemia and a similar marrow as described by METTIER et al (1930) in a third. He stated that there was no evidence of failure of maturation at any stage of development, such as distortion or failure of haemoglobinisation. In the two former cases the cells were simply fewer than usual. The marrow examination was repeated in the recovery phase and "presented the usual picture of response to loss of blood cells." He admitted that he found difficulty in eliminating failure of maturation but favoured PARSON and SMALLWOOD's view (1935) that the main causal factor was a slowing down of erythropoeisis.

The following year McMILLAN and INGLIS (1944) presented their results on six sternal marrow examinations. One was megaloblastic with achlorhydria. Two with normoblastic marrows, with a few megaloblasts present, had achlorhydria in one and a low free Hcl in the other. The other three had normoblastic marrows with free acid present in the fractional test meal. Unfortunately no follow up studies or comments were made.

VILTER et al (1946) published the bone marrow findings in their eleven cases. Five patients had moderately

hypercellular marrows, an equal number were normally cellular and the one remaining showed moderate hypocellularity. Differential counts showed a relative increase in the erythrocyte progenitors, with the majority of these cells at the normoblast or late erythroblast stages of development. In one severe case (17% packed cell volume) the bone marrow was megaloblastic.

The granulocyte series was normal.

All these abnormalities disappeared when vitamin C alone was administered.

In discussing their findings they feel that the bone marrow was never as hypercellular as one would expect in pure haemolytic anaemia or the anaemia of acute blood loss. Although they favour haemolysis as the cause of the anaemia of scurvy they admit that there are significant factors other than haemolysis in the etiology. "The conclusion seems inescapable that vitamin C reversed these abnormal processes and is essential for normal formation and maintenance of erythrocytes."

Unfortunately no remark is made with regard to mitosis in their cases.

The gastric analyses in their cases showed achlorhydria or hypochlorhydria after histamine stimulation.

Amongst the case records of Cases 14 to 32 there

are five reports on bone marrow examinations. Cases 24, 25 26, 28 and 29 all showed normoblastic hyperplasia but in one of these (Case 26) "numerous megaloblasts" were seen. This case was macrocytic as determined by the mean corpuscular volume.

With the exception of the non-anaemic case (Case 7) thirty bone marrow examinations were performed on the cases in the Personal series (Cases 1 to 13). In most cases an initial and a follow up study after vitamin C therapy was all that was considered necessary. In others (Cases 6, 8 and 9) subjected to prolonged control periods, several bone marrow examinations were performed during these periods. In this way it was possible to observe the influence of other haematinic drugs, such as folic acid, vitamin B_{12} and iron added separately to the diet on which the disease developed.

In all cases, except Cases 2 and 11, a similar picture was observed. Increased cellularity of the red cell series was the rule. This was more marked in the more severely anaemic cases. The hyperplasia was normoblastic in all but in three cases a few megaloblasts were observed. It was the impression that the severer the anaemia the further back in the red cell maturation the hyperplasia occurred. This impression is reflected in the differential bone marrow counts in Table 12. In this Table the differential white cell percentages have been omitted but the leucocyte:erythrocyte bone marrow ratio (L:E ratio) has been included. Consequently

the percentages of nucleated red cells in the table represent the percentage of the total number of all nucleated cells.

Table 12.

Case No.	V.P.C. %	L:E Ratio	Normoblasts				Megaloblasts %	Units Free Hcl. Histamine	
			Bas. %	Poly. %	Orth. %	Total %		Before	After
1.	14	0.41	22.4	20.2	18.8	60	8	0	0
4.	23	1.83	5.0	9.3	19.2	33.5	0.8	0	17
6.	15	1.3	9.0	17.0	16.6	42.6	0	0	0
8.	22	1.2	6.0	17.4	21.0	43.8	0	0	0
9.	23	1.05	9.2	17.4	19.0	45.6	0.6	0	0
10.	26	2.08	4.0	8.4	20.6	33.0	0	7	20
11.	42	2.3	3.5	5.5	22	31.0	0	10	
13.	15	1.5	5.6	8.2	21.2	35.0	2.2	15	11

This table serves to show that the more severe the anaemia the smaller is the L:E ratio, and the higher the percentage of the earlier red cells.

No significant alterations in the bone marrow picture occurred following the administration of vitamin B_{12}, folic acid or iron, but ascorbic acid rapidly reversed these abnormal changes.

Marrow examinations performed at the peak of the reticulocytosis showed a similar picture of hypercellularity as was present prior to treatment. Prior to treatment clumps or "nests" of basophilic or polychromatic normoblasts were often seen. At the peak of the reticulocytosis similar clumping was

observed. This similarity was only in the degree of cellularity, however. Mitotic figures were rarely seen in these "nests" prior to treatment. The mitotic stage in these cases, when seen, was in telophase which would represent the lag karyokinetic curve of decreased mitosis as described by LEITNER (1949). The appearance was, then, of numerous cells lying dormant. At the peak of the reticulocytosis the marrow presented the picture of intense activity. In each clump a mitotic figure was very frequently found with both prophase and metaphase stages predominating. These figures were more noticeable in the late normoblasts, whereas prior to treatment, the only mitotic figures seen were in basophilic normoblasts.

Marrow examinations performed two weeks after treatment were indistinguishable from normal, in all cases.

There can be no doubt that vitamin C was responsible for reverting these abnormal marrow changes to normal, as, at this time, the patients were still on the diet on which they developed the disease.

These marrow findings are similar in many respects to those described by METTIER and CHEW (1932) in the guinea-pig. Decreased cellularity as described by ISRAELS (1943) and VILTER et al (1946) was not seen. Vilter et al's case was 75 years old whilst Israel's two cases were not severely anaemic.

The hypercellularity seen in these cases, when

taken together with the decreased mitosis, may mean retarded erythropoeisis. As the anaemia progressed, this retardation made itself more obvious by an increasing number of the early red cell progenitors. A disproportionate slowing through the stages up to the release of mature erythrocytes into the general circulation, as opposed to the white cell development may account for the increased number of red cell precursors present. All cases except Cases 1 and 6 had a total white cell count over 6,000 at the time of marrow examination.

The prompt and rapid response to ascorbic acid would favour such a conclusion.

SUMMARY OF THE BONE MARROW FINDINGS IN ADULT SCURVY.

1. The more severely anaemic the patient, the more hypercellular the marrow appeared to be.

2. The L:E ratio decreased and the percentage of the earlier red cell precursors increased with the severer cases.

3. Despite this cellularity the marrow looked somewhat inactive, as only rarely were mitotic figures seen.

4. On the other hand marrow examinations at the peak of the reticulocytosis presented a picture of intense activity.

5. The administration of vitamin B complex, vitamin B_{12}, folic acid and/or iron in three cases had no effect on these marrow changes.

6. Ascorbic acid administration, however, in all cases, whether the above substances had been given or not, rapidly reverted these changes to normal.

MECHANISM OF THE ANAEMIA IN SCURVY.

Evidence for Intravascular Haemolysis.

Raised reticulocyte counts, increased urinary and faecal urobilinogen levels with no bile in the urine, and slight or moderate jaundice, made accelerated blood destruction seem likely to VILTER et al (1945)(1946) as the mechanism of the anaemia of Scurvy.

All these signs disappeared four to twelve days after vitamin C was administered.

No patient had clinical evidence of hepatic dysfunction. Eleven patients had twenty-nine liver function tests performed and only nine tests were definitely abnormal. Apart from the icteric index and qualitative van den Bergh reaction, albumen globulin ratios were done on only four patients, cephalin cholesterol flocculations in seven and bromsulfalein excretions in the same seven.

In the discussion after Vilter's first presentation of their cases [VILTER et al (1945)] C.J. Watson favoured hepatic dysfunction rather than haemolysis as the cause of these features.

It is well known that hepatic dysfunction may be present without any of the above tests being abnormal. The raised urinary urobilinogen levels, estimated semi-quantitatively by the serial dilution method of WALLACE and DIAMOND (1925)

would have to be explained. Faecal urobilinogen estimations were performed on three patients and were markedly elevated in two and slightly elevated in one.

The authors considered that the degree of anaemia and the degree of jaundice as judged by icteric indices and qualitative van den Bergh estimations could not be correlated with the extent of visible ecchymosis, and suggested that some haemolysis occurred intravascularly.

On their marrow findings, however, they suggested that there were significant etiologic factors other than haemolysis. The bone marrow was never as hyperplastic as one would have expected in haemolytic jaundice and the total white blood cell counts frequently were low.

Many years before MEYER and McCORMICK (1928) suggested that abnormal blood destruction might be responsible in the anaemia of scorbutic guinea-pigs but as METTIER and CHEW (1932) point out, convincing evidence of this is lacking.

In the personal series here presented one was struck with the many features that suggested haemolysis as a mechanism although the red cell fragility in saline was not increased and the white cell counts were low. Consequently it was decided to carry out accurate quantitative urinary and faecal urobilinogen studies because urobilinuria, as determined by the spectroscopic test, appeared to be an almost constant finding on admission.

Daily urobilinogen studies showed a gradual drop in urinary levels with the patients on the control diet and in bed, whereas this was not the case with regard to the faecal levels (See Fig. 21).(page 218).

During this period of observation it was noticed in the first case studied (Case 6), that the urinary urobilinogen began to rise, followed twenty-four hours later by a rise

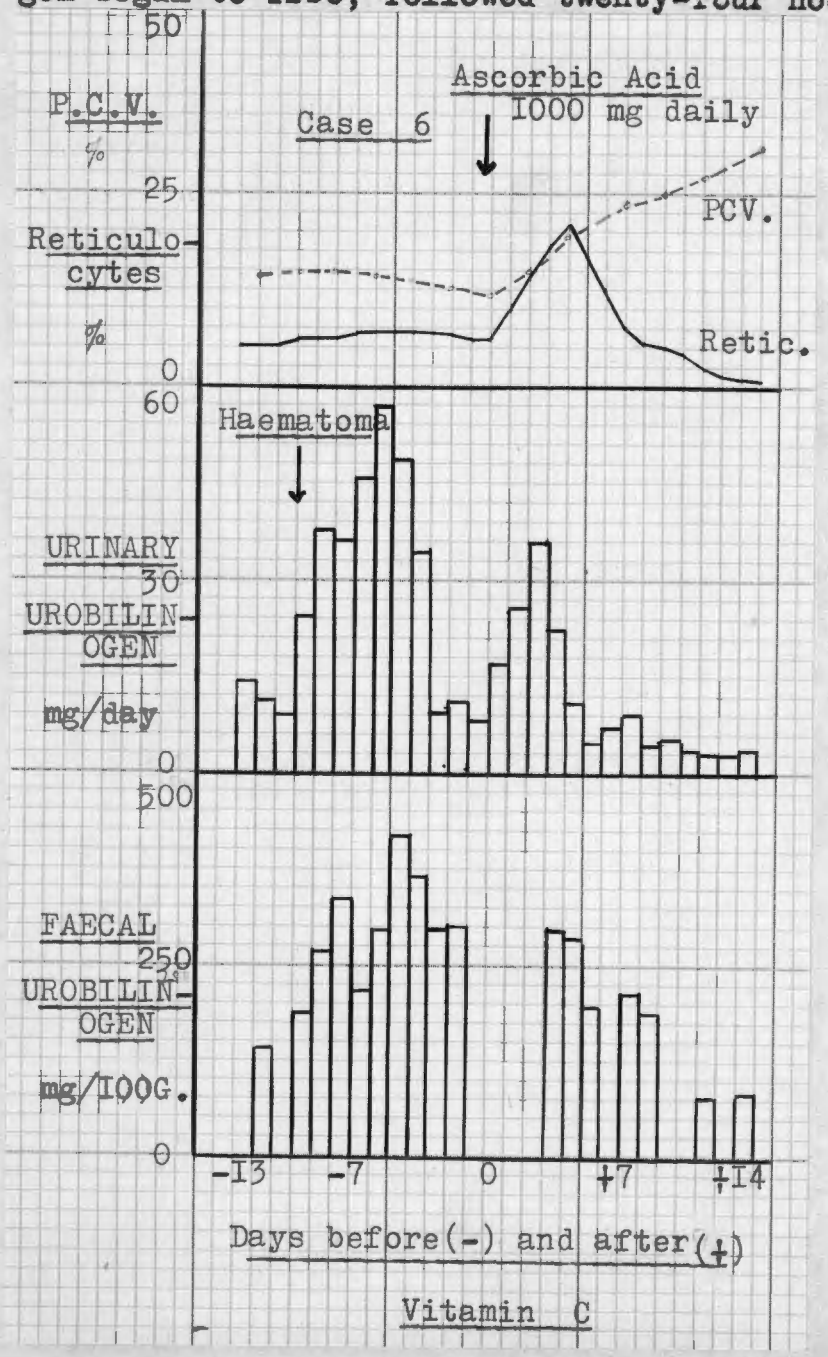


FIGURE 19

(Case 6.)

Note :- A rise in urinary and faecal urobilinogen levels followed accidental haematoma formation No change in the reticulocyte count or PCV. was associated.

in the faecal urobilinogen (See Fig. 19). At the time it was felt that a haemolytic crisis was occurring but no change in reticulocytes, packed cell volume or white blood cells took place.

The day prior to this rise, however, large haematomata had resulted from attempting a capillary fragility test.

A mild scorbutic (Case 7) with gross liver dysfunction, showing a marked urobilinogenuria, was in the ward during the same period. He served as an excellent control as he was not anaemic, had no deep haematomata and no raised faecal urobilinogen. Blood pressure cuffs were applied but no haematomata resulted consequently forty ml. of blood were removed from his vein and injected into his buttock. The urobilinogenuria immediately became more marked and there was a rise of faecal urobilinogen forty-eight hours later, although the latter was still within normal limits (See Fig. 20). Forty ml., however, is a very small amount of blood. When the urobilinogen levels had settled, an equal quantity (forty ml.) of blood was withdrawn and discarded to see whether any change occurred merely from the small venesection. No change occurred. That urobilinogen levels may fluctuate from day to day is well known, so convincing proof that the raised urobilinogen levels were merely due to extravascular haemolysis in the haematomata was, as yet, lacking.

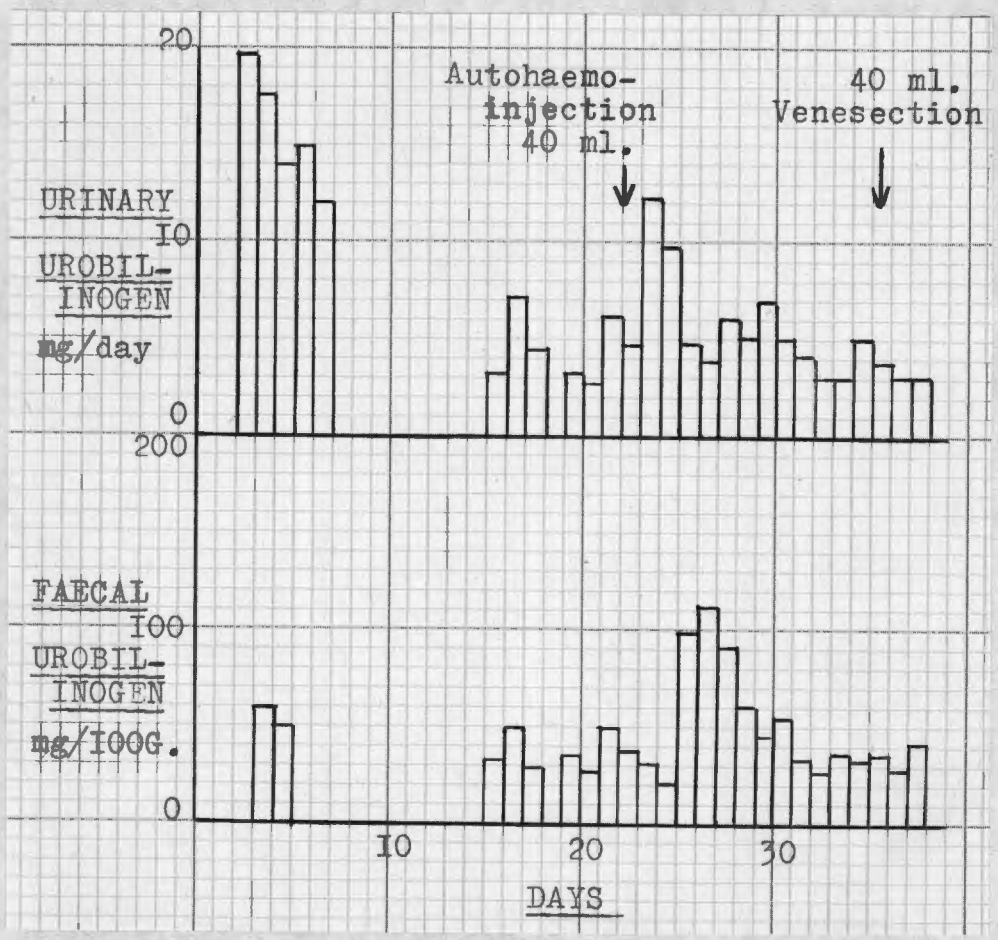


FIGURE 20 (Case 7)

The next case admitted (Case 8) gave this proof. His urinary urobilinogen fell slowly with rest in bed so that by the fifteenth day his urinary level was almost normal yet his faecal level was still abnormal. His large deep haematoma of the calf and popliteal fossa was slowly absorbing. The faecal urobilinogen gradually fell to within normal limits and the calf measurement decreased another ½-inch in circumference. The leg was no longer tender. A blood pressure

cuff was applied to the thigh. Pain, tenderness and an increase in size of the calf followed, with a faecal urobilinogen of over 1,000 mg. per 100 G. over the next four days. There was no change in the already elevated

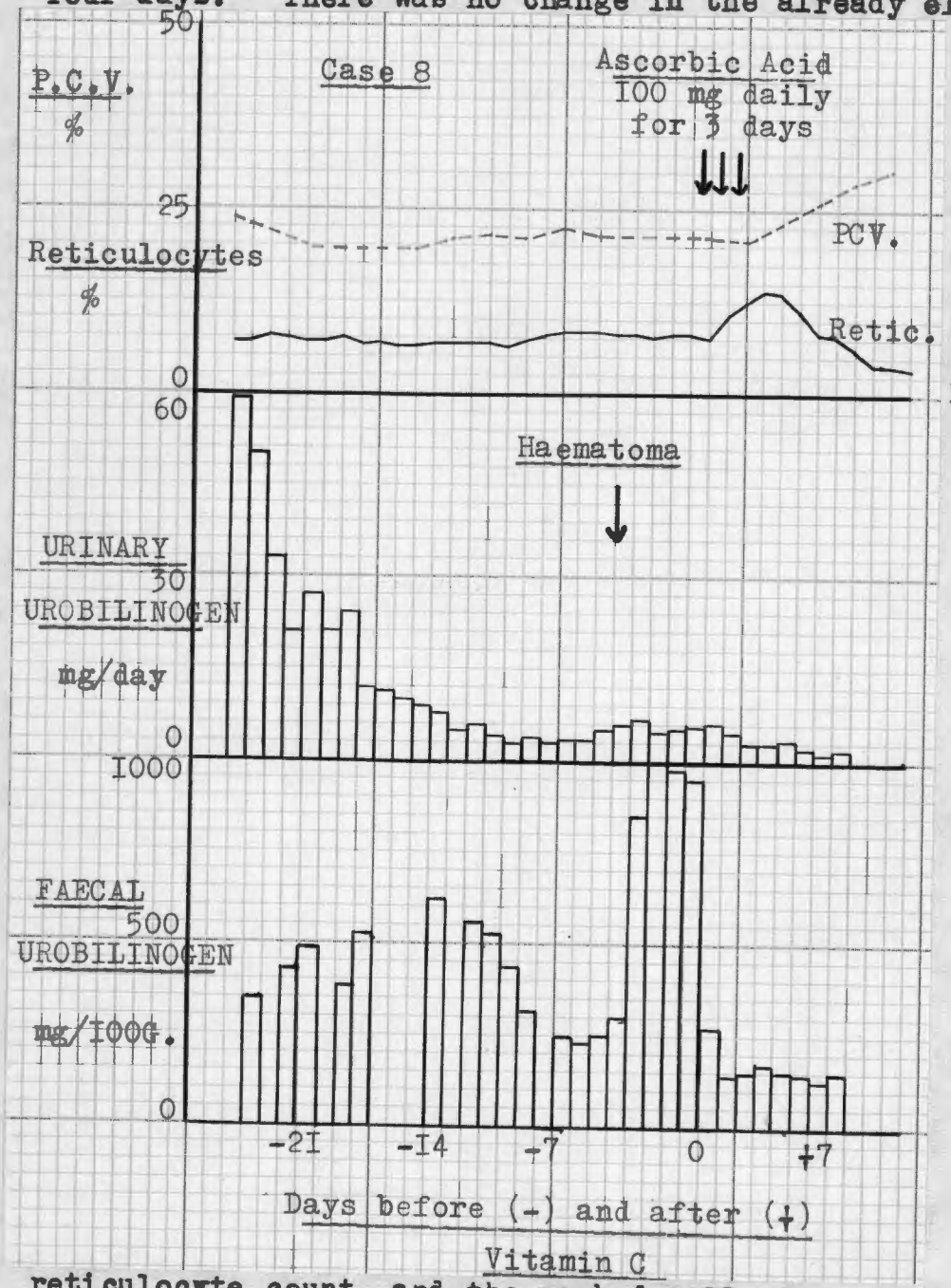


FIGURE 21

(Case 8)

"Deliberate" haematoma production.

reticulocyte count, and the packed cell volume remained at 22%. It was indeed interesting that unlike Cases 6 and 7,

there was no "spill over" in the urine. This would give a normal urine-stool urobilinogen ratio [STEIGMANN et al (1943) and WATSON (1937)] and thus suggest liver function able to deal adequately with excess urobilinogen presented.

MACLAGAN (1946) chose amongst others, fracture cases as normals for his method of faecal urobilinogen estimation. These cases soon had to be excluded as they had very high faecal urobilinogen levels. Unfortunately no urinary levels were done at the same time. The faecal levels recorded appeared in his opinion to be too high to result merely from the local haematoma formation. Likewise VILTER et al (1946) felt that the visible ecchymoses present could not account for the degree of haemolysis, judged by these methods.

WATSON (1937) noted a normal level of urinary urobilinogen in a fractured femur but did not perform a stool estimation. In another case of post-traumatic haematoma in the kidney, he noted a serum bilirubin of 0.3 mg. %, with a normal urinary urobilinogen but a faecal estimation of 1,100,000 mg. per day. Furthermore the icteric index of his patient was 29. "The elevated icteric index and the patient's colour with no bilirubinaemia were obviously due to the haematin present (readily identified by the haemochromogen spectrum)".

The "jaundiced" sera without a rise in serum

bilirubin was noted in the cases presented here. Icteric index readings of 22, 24 and 25 units with normal serum bilirubin levels were obtained but the presence of haematin was not investigated.

In a fourth patient (Case 9) in this series attempts at producing haematomata with the blood pressure cuff method failed. Apart from a fall in urinary urobilinogen levels, on bed rest, there were no marked fluctuations in the faecal urobilinogen content, reticulocytes or white cell count. The faecal urobilinogen reached normal levels just prior to treatment but at this time the packed cell volume had dropped to 17.5% from an initial level of 23%. At this time, too, his haematoma was only just palpable. Consequently this case establishes, again, a direct relationship between the high faecal urobilinogen levels and the extent of haematoma formation.

This is further reflected by the graph of Case 6 (See Fig.19) in that the faecal urobilinogen lagged far behind the urinary levels in returning to normal following vitamin C treatment. This severely anaemic case's rapid drop in urinary urobilinogen coincided with a rapid improvement in his anaemia after vitamin C was given. This graph also shows a second rise in urinary urobilinogen just prior to the peak of the reticulocytosis due to ascorbic acid therapy. Recent work[LONDON et al (1950)] has confirmed a view held by WHIPPLE et al (1930) that a considerable amount of the urobilinogen may result from the haemoglobin precursors rather than

haemoglobin breakdown. At the same time, however, the patient was constipated which, according to WATSON (1937), may lead to increased resorption of urobilinogen and account for this second rise.

The findings in other cases studied in this series from this aspect are illustrated in Table 13.

Table 13.

Case No.	P.C.V. %	Serum Bili-rubin mg.%	Urobilinogen		Haematoma	Liver Function Tests
			Urinary mg./day	Faecal mg./100 G.		
6.	13	0	57.0	402	++	Normal
7.	53	0.5	19.6	77	-	Abnormal
8.	22	0	59.9	427	++	Abnormal
9.	23	0	21.2	260	++	Abnormal
10.	26	0	15.8	303	++	Abnormal
11.	42	1	20.8	395	++	Abnormal
12.	27	0	13.5	163	++	Abnormal
13.	15	0	63	510	++	Abnormal

Within a week of bed rest the urinary levels had fallen from high initial levels to normal or near normal. The faecal urobilinogen remained elevated for a week to ten days after treatment in those given ascorbic acid soon after admission.

From this table and the graphs just discussed certain important facts emerge.

1. The faecal urobilinogen.

These levels appear to be related to the degree of haematoma formation as:-

(a) With rest in bed and slow resolution of the haematoma as judged by the decrease in its size and degree of tenderness, the faecal urobilinogen content as slowly decreases.

(b) Increasing the degree of haematoma formation leads to a rise in the faecal urobilinogen level.

(c) High faecal urobilinogen levels were found in a non-anaemic case with a large haematoma (Case 11) and, in a non-anaemic case without any haematomata (Case 7), low levels were observed.

(d) A rapid drop in faecal urobilinogen levels did not occur with treatment as one would expect from cessation of intravascular haemolysis. Furthermore neither would this observation fit the view that failure of utilisation of haemoglobin precursors accounted for the high faecal urobilinogen levels present.

(e) Fluctuations in the faecal urobilinogen levels did not coincide with changes in packed cell volume, reticulocyte count or white cell count. Whilst in those cases where the anaemia grew worse on bed rest alone, a decrease in the faecal urobili-

nogen occurred, provided that no fresh haematomata were produced.

2. The urinary urobilinogen.

The presence of urobilinogen in the urine appears to depend upon the adequacy of the liver function as:-

(a) High levels occur in non-anaemic scorbutics with (Case 11) or without (Case 7) haematomata.

(b) The high level dropped to normal or nearly normal levels in the control period as the liver function improved with bed rest [WATSON (1937)].

(c) Producing a rise in the faecal urobilinogen by causing further extravascular haemolysis, does not necessarily cause a rise in the urinary level.

(d) In the severely anaemic case (Case 6) with a high urinary urobilinogen level, a rapid drop in this urobilinogen content after vitamin C was given, coincided with a rapid improvement in the anaemia with its attendant anoxaemic effects.

It is unlikely, then, from the foregoing statements that the anaemia of scurvy results from intravascular haemolysis. Extravascular haemolysis, likewise, cannot account for the anaemia. It would serve to explain, however,

the excess urobilinogen excretions in scurvy, and the raised icteric indices in the absence of bilirubinaemia. The effect of this extravascular haemolysis also precludes the use of these pigmemt studies as a means to determine further the mechanism of the anaemia. It would, however, not serve to explain the reticulocytosis that occurs prior to treatment with ascorbic acid.

Evidence for a Post-haemorrhagic mechanism.

JANET VAUGHAN (1934) suggested that the reticulocytosis seen prior to treatment could be due to the stimulation of the bone marrow by small repeated haemorrhages. WINTROBE (1946) states that blood regeneration following the administration of ascorbic acid does not prove that this vitamin is necessary for haematopoeisis. Cessation of haemorrhage and resorption of blood should permit restoration of normal blood values.

The bleeding tendency in scurvy occurs usually prior to the development of anaemia. Bleeding from mucous membranes and into the subcutaneous and intramuscular tissues could provide a basis for chronic blood loss with acute blood loss being superimposed every now and then.

The reticulocytosis occurring prior to treatment would mean that the bone marrow is unable to keep pace with the extent of blood loss, until vitamin C was available to prevent the continuation of this loss of blood.

PARSONS et al (1933)(1935) mention that in chronic cases of scurvy associated with large haemorrhages into the tissues and from mucous membranes a post-haemorrhagic blood picture becomes superimposed and the anaemia may then become hypochromic and in extreme cases even microcytic. At the same time the picture presented was, in their opinion, unlike that which arises from sudden loss of blood. In their cases

this hypochromic anaemia did not respond to iron. On the other hand KENNEY and RAPOPORT (1939) found that iron was necessary in two of their five scorbutic anaemic infants.

Hypochromic anaemia has been found in the scorbutic guinea-pig [ARON (1939)] but METTIER and CHEW (1932) in port mortem studies could not demonstrate that sufficiently large haemorrhages had occurred in various tissues to be responsible for the severe degree of anaemia that appeared in most instances. ARON (1939) agreed that the anaemia was not due to loss of blood as was formerly believed.

The same confusion exists in man. However most authors agree that the morphology and clinical picture is unlike that of acute or chronic blood loss.

McMILLAN and INGLIS (1944) found simple microcytic anaemia in fourteen patients and a microcytic hypochromic blood picture in another six. Their criteria for cell size were not based on either the haematocrit or mean cell diameter measurements. They conclude that iron lack was not a factor and in their forty cases the anaemia present bore no constant relation to the extent of haemorrhages.

On the other hand BARNES (1947) in reporting a case with severe anaemia, was of the opinion that haemorrhage into the legs was the cause of the anaemia. His conclusion was based on a bilirubinaemia of 2.1 mg. % and the satisfactory recovery without haematinic drugs, the blood "being reabsorbed and refashioned". On a sterilized milk diet with 20 mg.

ascorbic acid daily a rapid haematological response followed. Unfortunately no preliminary period of control to gauge the effect of bed rest only was adopted.

Reports exist, however, of failure of blood transfusion to correct the anaemia. JENNINGS and GLAZEBROOK (1938) in one case noted benefit lasting only ten days; previous to this iron had been ineffective. Furthermore it was their opinion that anaemia had been present a very long time before subcutaneous or other haemorrhages were manifest. Likewise HATHERLEY (1947) reports the failure of thirteen pints of blood to change the haemoglobin level until vitamin C was given.

On the other hand artificially induced post haemorrhagic states both acute and chronic [CRANDON et al (1940), LOZNER (1941)] by venesection of 500 ml. to 1600 ml. at a time, or by repeated withdrawal of small amounts over a prolonged period in scorbutics, have led to spontaneous regeneration in one [CRANDON et al (1940)] and in another a failure of spontaneous regeneration until vitamin C was given [LOZNER (1941)]. RALLI and SHERRY (1941) maintained a mild scorbutic with a recent history on a diet deficient in vitamin C over 52 days, and despite fresh bleeding from frequent catheterisation and a reappearance of ecchymosis and bleeding from the gums, no drop in either the haemoglobin or red cells occurred.

VILTER et al (1945) and SHULTZER (1936) noted how bed rest prevented further haemorrhages from occurring,

SHULTZER (1936) even inducing relapses by allowing the patients up. VILTER et al (1946) felt that the extent of the ecchymosis in their cases could not be correlated with the anaemia present. Gums oozed with slightest trauma but no case lost more than a few ml. of blood daily. No increase in ecchymosis occurred after the patients were hospitalised yet the anaemia grew rapidly worse in some cases.

In the series presented here, only one case (Case 5) had any recent story of external blood loss. An epistaxis, apparently not severe, had brought the patient to hospital. No further bleeding occurred once he was placed in bed. No haematomata were palpable. He was the only case with a leucocytosis on admission and this fell to a normal value by the fifth day. With bed rest only and on the diet on which the disease developed the reticulocytes rose from normal levels to 7% but fell gradually to 3.5% by the tenth day. After a drop by 1% two days after admission, the packed cell volume climbed 4% over the next six days, yet three days later the packed cell volume was still at the same level (See Fig. 17 - Chapter 6). Ascorbic acid was then given. By the fifth day a reticulocyte peak of 15% was reached and the dramatic response which followed cannot but be significant when the 12½% rise in packed cell volume over the ten days following treatment is compared with the 4% rise over the ten days preceding treatment.

Only seven of the thirty-two patients complained of bleeding gums, despite the fact that gum changes were present

in all. No case stated that more than a few ml. of blood was lost a day. Once in bed the cases in the personal series did not ooze from their gums at all. Furthermore the large haematomata present in some actually decreased in size and even disappeared, yet over a period on the control diet and bed rest over twenty-five and thirty days the anaemia grew worse in the three selected for study.

Mention has been made that by raising the capillary pressure, fresh bleeding into the tissues was produced, but no change in reticulocyte count or drop in packed cell volume followed. It would seem unlikely, then, that the reticulocytosis seen prior to treatment in these cases was due to repeated stimulation of the bone marrow by blood loss.

Detailed morphological studies show that the blood picture is most unlike that of blood loss.

In acute and chronic blood loss the capacity of the serum to bind iron is increased [CARTWRIGHT and WINTROBE (1949)]. In the cases studied here, the iron binding capacity was markedly reduced.

Confirmation of the failure of iron to alter the anaemic state in scurvy, as was found by other investigators, was shown by a failure of response to large doses administered intravenously in three cases, prior to ascorbic acid therapy (Cases 6, 9 and 13), and a failure to cause a "double reticulocyte response" or cause further regeneration after an

initial response to smaller doses of ascorbic acid had occurred (Case 8).

On the diet on which the disease developed, without preceding or concomitant iron therapy ascorbic acid was able to produce within three to four weeks complete haematological regeneration.

Finally, it was observed in this series, as did others, that the severity of the anaemia could not be correlated with the degree or extent of haematoma formation or external blood loss.

CONCLUSIONS.

1. It has been shown that the anaemia of scurvy is not merely due to loss of blood, as occurs in other bleeding diseases such as purpura haemorrhagica and haemophilia.

2. Loss of 500 ml. or more of blood experimentally in mild scurvy will not induce anaemia, but in severer degrees of vitamin C lack anaemia results, which fails to regenerate until vitamin C is given.

3. Bed rest decreases the occurrence of haemorrhage both externally and into the tissues, yet the anaemia may get worse. Increasing the size and tenderness in a haematoma or producing further

haematomata by constriction proximally leads to no change in reticulocytes or packed cell volume.

4. No correlation exists between the extent of haematoma formation or external blood loss and the degree of anaemia.

5. Finally, the anaemia of adult scurvy resembles in no way the anaemia of chronic blood loss in its morphology, in the capacity of the serum to bind iron, or in its response to iron therapy.

THE PROBLEM OF DYSHAEMOPOEISIS IN SCURVY.

The overwhelming weight of the evidence, so far submitted, points to the anaemia in scurvy being dependent upon the effect on the erythron, of a chronic lack of vitamin C in the body. How this effect is produced has not yet been discussed. SHAFAR (1949) states that ascorbic acid may possibly participate in the various enzyme reactions of the organism. In such a way the dyshaemopoeisis resulting may be due to a secondary effect of vitamin C lack, i.e. the absence of vitamin C may interfere with the metabolism of the factors necessary for red cell production. On the other hand, vitamin C itself may be primarily responsible for red cell production.

This mechanism is usually classified into two sections:

1. Deficiency dyshaemopoeisis
2. Toxic dyshaemopoeisis.

The latter will be considered first.

A. Chemical Substances.

HUGHES (1950) has shown the close similarity of certain clinical syndromes of metallic poisoning to certain clinical deficiency patterns. Such substances may cause normocytic anaemias with bone marrow hypofunction. These metals may act as "anti-vitamins" in the way arsenic attacks the sulphur grouping of the protein to which vitamin B_1 is linked as part of an enzyme system. With respect to scurvy

the close clinical similarity to the effects of benzene poisoning has been noted [British Medical Journal (1949)]. Signs of vitamin C deficiency have developed in men exposed to benzene fumes. Ascorbic acid in large amounts given with its oxidase, which is present in cucumber, protects guinea-pigs against benzene vapour.

The nature of the occupations followed by the patients in this series did not allow any exposure to such industrial hazards.

B. Infection.

Mention has been made that infection increases the metabolic requirement of vitamin C. On the other hand particularly in guinea-pigs and infants scurvy may be complicated by infection. In man RALLI and SHERRY (1941) have recorded abscess formation in a haematoma and the relation between the gum changes in scurvy and dental infection has been noted. Pyrexia and local signs of inflammation may accompany the haematoma of scurvy to the extent that frequently cellulitis and osteitis have been erroneously diagnosed.

The anaemia accompanying infection is usually normocytic and normochromic. Recent work [FINCH (1948), CARTWRIGHT and WINTROBE (1949) and TOTTERMAN (1949)] shows that amongst other features, a low plasma iron level is constantly found. Together with this, the iron binding capacity of the serum is low. Experimentally the above features with anaemia

may be produced by causing a sterile abscess from the intramuscular injection of turpentine [CARTWRIGHT and WINTROBE (1949)]. Resolution of the anaemia occurs only when the infection resolves.

In scurvy a low plasma iron has been reported in guinea-pigs [BRAGANCA and SAHA (1943)]. SCHRODER and BRAUN-STAPPENBECK (1941) are quoted by TOTTERMAN (1949) as finding the serum iron levels increasing parallel with the serum ascorbic acid levels as vitamin C was given to a case of scurvy. On the other hand normal values were found in three scorbutics studied by VILTER et al (1946).

ALBERS (1943) quoted by SHAFAR (1949) presented evidence that the intravenous injection of ascorbic acid increased the concentration of serum iron in infections. Such results in the anaemia of infection could not be confirmed by the extensive studies of TOTTERMAN (1949) even with prolonged large doses of ascorbic acid orally or parenterally.

Unfortunately no reports on the iron binding capacity of the serum in scurvy were available for study.

In seven patients presented in this series (Cases 6, 8, 9, 10, 11, 12 and 13) the plasma iron determinations were all low. Brief mention only will be made of the results as full details will be published by Dr. O. Budtz-Olsen who kindly performed these tests for me.

In each case an identical picture resulted.

Prior to treatment with ascorbic acid, whether previous intravenous iron therapy, other haematinic or antibiotic (In Case 9) had been given or not, the plasma iron levels were very low, in the region of 20 to 40 u G. per 100 ml. Five minutes after an intravenous injection of iron the level was only in the region of 150 u G. instead of the normal total iron binding capacity of 300 to 400 u G. per 100 ml. After ascorbic acid therapy only, i.e. with the patient still on the control diet, improvement in both the plasma iron level and total iron binding capacity occurred.

There is no doubt, then, that the relation to the anaemia of infection goes far deeper than it superficially appears. In support of this is the fact that, despite the low plasma iron levels, neither the anaemia of scurvy nor the anaemia of infection responds to iron administered orally or parenterally. Whether the mechanism of production of the two anaemias is the same, however, is quite another matter.

That the low plasma iron levels in scurvy may be merely incidental and not a factor in the pathogenesis of the anaemia of scurvy is suggested by Case 11. His plasma iron and iron binding capacity was one of the lowest found (28 u G. per 100 ml. and five minutes after 50 mg. of iron as "Ferrivennin" intravenously the level was only 54 u G. per 100 ml.). The packed cell volume was 42%. The outstanding feature common to all seven patients was the presence of one or more intramuscular haematomata. This, when the experimental turpentine abscess is taken into account, may provide the

common basis of the relation to the anaemia of infection. Further evidence in favour of this was a drop in plasma iron from 44 u G. per 100 ml. to 18 u G. per 100 ml. in a week in the control period before treatment with ascorbic acid (Case 8). Just before the second reading was taken, further bleeding into the haematoma had been produced with proximal constriction using a blood pressure cuff. The packed cell volume remained at the same level (22%).

Unfortunately no anaemic case of scurvy without haematomata was available for study at this time. On the above hypothesis, normal plasma iron levels should be found. No large intramuscular haematomata were mentioned in the two available case reports of the three normal plasma iron levels reported by VILTER et al (1946). The lowest level reported was 72 u G. per 100 ml. He had "large deep purpuric" spots but no ecchymosis. He was classed as a severe scorbutic. Two milder cases had levels of 116 u G. and 83 u G. per 100 ml., respectively. The former had extensive popliteal and antecubital ecchymoses and the case report of the latter was not available.

Proof that this low plasma iron and iron binding capacity has anything to do with the pathogenesis of the anaemia of infection, is as yet lacking. It is suggested that the hypoferraemia is due to iron transferred to the reticulo-endothelial system [TOTTERMAN (1949)]. It is of interest to note here that BRAGANCA and SAHA (1943) found no

significant increase in the liver iron in anaemic scorbutic guinea-pigs. Furthermore bringing the serum iron to normal levels with prolonged parenteral iron therapy had no effect on the haemoglobin levels or red cell count [TOTTERMAN (1949)].

A considerable difference, however, between the anaemia of scurvy and the anaemia of infection exists in their response to ascorbic acid therapy. Prolonged large doses of ascorbic acid administered orally or parenterally, has no effect on the anaemia of infection or the plasma iron level even when given together with iron. [TOTTERMAN (1949)].

Not only does the anaemia in scurvy rapidly improve on adding only ascorbic acid to the diet on which the disease developed, but the plasma iron levels also improve.

It would appear, then, that undetected concomitant infection in scurvy is not responsible for the associated anaemia.

The fact, that a sterile intramuscular abscess produced experimentally may lead to anaemia, could by analogy, lead to the consideration of the possible role played by the intramuscular haematoma in the anaemia of scurvy. Certainly the anaemia in scurvy usually develops after the deeper haemorrhagic manifestations appear. Furthermore it is possible that diffuse haemorrhagic effusions may lie along the deeper fascial planes without clinical evidence of their presence. Consequently to state that anaemia may occur in

scurvy without the presence of these deeper haemorrhagic manifestations, may be incorrect if clinical means are used in such judgement.

In those cases in this series studied over a prolonged control period, resolution of the large palpable haematomata, with loss of the local signs of inflammation and a decrease in their size, did not lead to resolution of the anaemia. Increasing the degree and extent of the haematoma formation (Cases 6 and 8) with the attendant pain and development of other local signs of inflammation, did not lead to deterioration of the anaemic state. The auto-haemo injection of 40 ml. of blood intramuscularly, likewise, did not cause the development of anaemia in a non-anaemic scorbutic (Case 7).

The response to ascorbic acid therapy was prompt and was complete often long before resolution of the haematoma occurred (Cases 3, 4, 6, 10 and 12).

Evidence, then, for dyshaemopoeisis from vitamin C lack, due to associated toxic factors is not convincing.

C. <u>The Part Played by the Thyroid</u>.

PARSONS and SMALLWOOD (1935) compare the morphology of the anaemia of scurvy with that of cretinism, i.e. "occasionally macrocytic, usually normocytic and never microcytic".

In this respect it is of interest to note that hyperthyroidism increases the demands for vitamin C [SHAFAR (1949)] and that CRANDON et al (1940) noted a drop in the basal metabolic rate during his phase of clinical scurvy. This was considered to be insignificant, however, as no rise or return to normal followed immediately after vitamin C therapy.

In this series no signs of thyroid dysfunction were encountered.

D. The Question of Liver Dysfunction.

In Chapter III reference was made that in almost every case in this series, some evidence of hepatic dysfunction was present. It may be argued that this may be the factor determining the onset of the anaemia in scurvy. High faecal urobilinogen and urinary urobilinogen levels often with reticulocytosis made WATSON (1937) feel that haemolysis was the underlying factor.* This view is not now generally held [British Medical Journal (1950)]. Macrocytosis seen frequently in liver disease has been suggested as due to the flat cells which are usually present, leading to a diameter thickness ratio of more than normal. RATH and FINCH (1949) found normal plasma iron and iron binding capacity levels. Consequently the morphological characteristics of the anaemia of scurvy are most unlike those associated with the anaemia of severe liver disease. Liver dysfunction as measured by the

*i.e. in the anaemia of liver disease.

liver function tests was as common in scorbutics without anaemia, as in those with anaemia. It would be difficult to consider liver disease as a factor in the pathogenesis of scorbutic anaemia when one considers the prompt and complete response to ascorbic acid without any other alteration in the management of the case. Liver function improves on bed rest [WATSON (1937)] and this was shown in these cases by a decreasing urinary urobilinogen excretion. Improvement in the anaemia did not necessarily follow.

It would be more difficult to rule out liver dysfunction as the cause of the reticulocytosis in these cases. No increased reticulocyte levels were present in those with milder degrees of anaemia (Cases 2, 7 and 12) and increased reticulocyte counts, although always present after a few days of bed rest, were absent on admission in most of the remaining, more severely anaemic cases. Liver dysfunction, as measured by the chemical tests, was as severe in the former as the latter.

It was felt that a more likely explanation for the increased reticulocyte levels in the anaemia of scurvy, could be found.

E. Deficiency of Essential Amino Acids.

In Chapter III the conclusion reached, with regard to liver dysfunction, was that the probable primary mechanism

was the deficiency of essential amino acids in the original diet. Mention was also made that the position was not clear as to what part, if any, vitamin C played in the metabolism of amino acids. On the one hand feeding tyrosine to scorbutic guinea-pigs [SEALOCK et al (1941)(1948)] to scorbutic infants [MORRIS et al (1950)] and to scorbutic adults [ROGERS and GARDNER (1949)] leads to a marked "tyrosyluria" which is abolished when vitamin C is given parenterally or orally. On the other hand MORRIS et al (1950) felt that failure of utilisation of essential amino-acids did not occur, as no stunting in growth or other features suggestive of malnutrition resulted in their infants.

Radioactive glycine studies have shown the importance of this substance in the synthesis of protoporphyrin [CARTWRIGHT (1947) LONDON et al (1950)] but the ease of synthesis of haemin makes the possibility that anaemia could result from a deficiency of this factor very remote [CARTWRIGHT (1947), FINCH (1948)].

On the other hand globin probably contains all the ten essential amino acids and a deficiency of these has produced anaemia in animals. This anaemia is usually normocytic and normochromic with a hypofunctioning bone marrow and a normal serum iron level [CARTWRIGHT (1947)]. It is possible that the haemin not utilised, due to this deficiency of globin, leads to increase bile pigment excretion [LONDON et al (1950)].

Although it was felt that the liver dysfunction in these cases probably resulted from a deficiency in the diet of amino acids no case showed hypoproteinaemia. Bile pigment studies showed increased excretion, but evidence has been presented to suggest that this originates from the extravascular haemolysis in the haematoma. At the same time it would be expected that the protein from the degradation products in these haematomata would still be available for resynthesis. In guinea-pigs BRAGANCA and SAHA (1943) found no significant variation in the excretion of haemin iron in the normal, the prescorbutic or the scorbutic state. Furthermore there was no significant difference between the liver iron of the scorbutic guinea-pigs and normals. Definite proof, however, that haemin occurs in excess when there is a deficiency of globin is, as yet, lacking. Further difficulty in evaluating the part played by amino acids is encountered if one considers that vitamin C, by virtue of its physiological properties, may be essential for amino acid metabolism, as the work on tyrosine in scurvy may suggest.

Significant in the whole problem, however, is the prompt and complete response to ascorbic acid, added to the diet on which the disease developed. This occurred without any amino acid or other factor being necessary before, during or after the addition of ascorbic acid. It would appear, then, that a dietary deficiency of amino acids played no part in the pathogenesis of the anaemia of scurvy.

F. Deficiency of the Anti-anaemia Principle and Other Vitamins.

In this section are included those factors necessary for the normal maturation of the red cell connected with the anti pernicious anaemia principle of liver, e.g. vitamin B_{12}, folic acid and other vitamins of the B group - the "Haemopoeitic vitamins". These anaemias are usually macrocytic and the nutritional deficiency may result from a defective diet, or gastro-intestinal dysfunction.

Differentiation of these anaemias from the anaemia in scurvy from the morphological aspect, is exceedingly difficult. The similar cell size, appearance of the peripheral smear, leucopaenia together with histamine-fast achlorhydria and a painful mouth may lead to an erroneous diagnosis of pernicious anaemia or nutritional macrocytic anaemia. The occasionally associated megaloblastic bone marrow [JENNINGS and GLAZEBROOK (1938), McMILLAN and INGLIS (1944), VILTER et al (1946)] makes the relationship even closer.

In fact McMILLAN and INGLIS (1944) and VILTER et al (1946) attribute the anaemia of scurvy to a complex deficiency. The latter authors suggest that "a patient with severe vitamin C depletion may have no anaemia, until additional strain is placed on the bone marrow by a deficiency of extrinsic factor, protein, iron or other factors".

In support of this VILTER et al (1950) quote

MAY et al's (1950) work on monkeys, fed a diet deficient in vitamin C and folic acid. This led to a megaloblastic anaemia. Folic acid and not vitamin B_{12} produced a rapid reversion of the bone marrow to a more normal state. Ascorbic acid produced similar but less dramatic changes and did not prevent death of some of the animals. By analogy these authors believed that the macrocytic anaemia of infancy is caused by defective utilisation, potentiated by vitamin C deficiency, of the small amounts of folic acid available in the unsupplemented milk diets. For clinical support Vilter quotes the work on twenty-six infants with macrocytic anaemia by LUHBY and WHEELER (1949) who discussed the failure of vitamin B_{12} and the metabolic role of folic acid and vitamin C. Applying this relationship to scurvy, Vilter felt that probably these factors (folic acid and vitamin B_{12}) condition the appearance of the normocytic or slightly macrocytic anaemia of adult scurvy.

This relationship becomes even closer with the more recent work on "tyrosyluria" found after feeding tyrosine to scorbutics. WOODRUFF and DARBY (1948) state that phenolic compounds are increased in the urine of patients with untreated pernicious anaemia, and liver suspensions from folic acid deficient rats are better able to oxidise tyrosine after the addition of folic acid. Folic acid given to tyrosine fed scorbutics will not prevent scurvy but will considerably modify the induced "tyrosyluria" [SEALOCK and LEPOW (1948), WOODRUFF et al (1949)]. Parenteral vitamin B_{12} had no such effect.

A similar effect could not be produced in adult scurvy [VILTER et al (1950)]. MORRIS et al (1950) showed that no relation existed between the degree of ascorbic acid depletion and the promptitude and magnitude of this induced "tyrosyluria". Furthermore intramuscularly administered folic acid did not inhibit the "tyrosyluria" already established, but did inhibit the "tyrosyluria" when given concurrently with the tyrosine. On the other hand the "tyrosyluria", whether established or not, was inhibited by ascorbic acid. It was their feeling that these results do not necessarily imply a more complete utilisation of tyrosine and phenylalanine. Folic acid and ascorbic acid may rupture the aromatic nucleus and lead to non-phenolic residues in the urine.

An approach to the part played by these "haemopoeitic vitamins" in the anaemia of scurvy may lie along the lines of the therapeutic effect of these vitamins. Quite rightly WOODRUFF et al (1949) point out that this anaemia often improves on hospital diets low in vitamin C. Reference, however, has already been made that, due to the very low minimal daily requirement of vitamin C and the effect of bed rest in diminishing this requirement further, these diets should be almost devoid of vitamin C before any conclusions can be drawn. On the other hand a megaloblastic anaemia in scurvy [JENNINGS and GLAZEBROOK (1938)] failed to respond to the ordinary hospital diet with liver supplements. METTIER et al (1930) also failed to induce a response with vitamin C free liver. There is always the difficulty that liver

preparations could contain vitamin C and other factors. With vitamin B_{12} and folic acid this possibility has largely been excluded. In three cases (Cases 6, 8 and 9) of this series vitamin B_{12} and folic acid were administered during the control period with a sufficient interval between to observe any response. No such response occurred in any case. In fact in two of the three cases the anaemia became more severe. Bone marrow examinations, repeated after these factors were given, showed no change. Furthermore all cases responded completely to ascorbic acid therapy and the rapidity of this response did not depend on a previous supply of these haemopoeitic vitamins. The effectiveness of smaller doses of vitamin B_{12} and folic acid, than those used here, in pernicious anaemia, tropical macrocytic anaemia, sprue, etc., has been convincingly shown [UNGLEY (1949), SPIES et al (1948), PATEL (1948), SUNDARAM (1948)]. These results suggest, then, that a deficiency of these factors in the diet did not play a part in the pathogenesis of the anaemia of scurvy.

Similar remarks may apply to the part played by other vitamins of the B complex group. This was suggested by LOZNER (1941) as a mechanism of cure in METTIER et al's (1930) series, i.e. that present in the orange juice and fresh liver used. CARTWRIGHT (1947) states that although deficiency of the components of the vitamin B complex may lead to anaemia in animals, it has yet to be produced in man. In this series very few signs of associated vitamin B complex deficiency were found and in four cases given vitamin B complex

in the control period no haematological remission occurred.

G. The Role of Iron in the Anaemia of Scurvy.

Ever since the discovery of vitamin C, its function in the organism has been connected with many phases of both exogenous and endogenous iron metabolism. ABT and FARMER (1938) could not produce anaemia in young scorbutic guinea-pigs on an adequate iron intake. Others, however, have produced the anaemia despite an adequate iron intake orally [METTIER (1938)] or parenterally [McFARLANE (1936)]. The associated achlorhydria, a diet deficient in all respects, and the occasional response to iron in infantile scurvy [KENNEY and RAPOPORT (1939)] have all helped to champion the cause of iron deficiency. MOORE et al (1939) claim that ascorbic acid given in conjunction with oral ferric salts causes a greater rise in the serum iron curve than when ferric salts are given alone. This together with the frequent achlorhydria has raised the problem of defective iron absorption in scurvy.

Several authors have reported the failure of iron therapy in scurvy [METTIER et al (1930), PARSONS and SMALLWOOD (1935), JENNINGS and GLAZEBROOK (1938), McMILLAN and INGLIS (1944)]. The oral route of administration was used, however. The only successful result with oral iron has been in a mild scorbutic made anaemic by venesection [LOZNER (1941)]. On the other hand the same procedure in a more severely scorbutic adult did not cause a response, until vitamin C was given [LOZNER (1941)].

In the discussion on the question of a post-haemorrhagic mechanism in scurvy it was stated that the morphology of the anaemia of scurvy resembles, in no way, the anaemia of iron deficiency.

The readily available iron in the haematomata and ecchymoses of scurvy should counter any possible dietary inadequacy. SUNDARAM (1948) states that "A primary dietetic deficiency of iron should not be postulated in adult males, unless every conceivable cause of excessive blood loss .....".

A low plasma iron level has led to speculation on the effect of vitamin C in endogenous iron metabolism. BRAGANCA and SAHA (1943) studied this in scorbutic guinea-pigs. They found no increase in the liver iron and despite the low food intake and the consequent low iron intake in the advanced stages of scurvy, the guinea-pigs were found to be in iron balance.

In an extensive study TOTTERMAN (1949) showed that no relation existed between vitamin C and iron physiologically or in the anaemia accompanying infection, despite the low plasma iron in the latter.

Furthermore a significant difference exists between the anaemia of scurvy and iron deficiency anaemia in this respect. Although both have low plasma iron levels, the iron binding capacity in the anaemia of scurvy is low, a state quite the opposite of iron deficiency anaemia.

The problem was tackled in this series from the aspect of therapeutic response.

(1) It was shown that iron was not necessary for the production of complete haematological remission either given before, during or after ascorbic acid therapy, in seven cases.

(2) Iron given intravenously in large quantities, to ensure that no absorption defect could participate, caused no response in the control period prior to ascorbic acid therapy in the three cases studied.

(3) In one case, an attempt was made to see if iron, given in large doses intravenously, could produce a "double reticulocyte" response after a preliminary small dose of ascorbic acid. No such response occurred. The original rapid response to the small dose of ascorbic acid slowed down and full regeneration did not occur until further ascorbic acid was given (Case 8).

Evidence then, that the lack of iron is instrumental in the development of the anaemia of scurvy, is without support.

H. Ascorbic Acid itself.

The specific effect of ascorbic acid on the anaemia of scurvy has been discussed and shown graphically. Evidence is submitted that anaemia is a late feature of scurvy

and that the absence of anaemia, seen in some cases of scurvy, appears to be a result merely of the degree of vitamin C lack. Vitamin C is necessary for many physiological functions in the organism [SHAFAR (1949)]. By decreasing the need in some functions, e.g. by placing the patient in bed, more vitamin C may be made available for other functions not related to locomotion.

The inconstant level of the reticulocytes on admission as compared with the constantly elevated level soon after the patients were placed in bed, would suggest that some endogenous source of vitamin C was stimulating the bone marrow. This source, however, was insufficient to cause further regeneration in many cases, a state analogous to the slight reticulocytosis without consequent haematological remission found when an inadequate dose of liver is given in pernicious anaemia [MINOT and CASTLE (1935)]. In other cases, usually where the duration of vitamin C lack is short or where the scurvy, as judged clinically, is mild, i.e. a state where the body stores would be more adequate, this preliminary reticulocytosis is followed by complete haematological remission on bed rest alone. If such a state of endogenous supply of vitamin C from the body stores was not responsible, it would be difficult to explain the slow remission of clinical features and the absence of progression of the disease on bed rest. This, judged by such clinical standards, as the tendency to bleed, etc., was noted not only in this series but by SHULTZER (1936) and others [VILTER et al

(1946), FOX (1940)]. Furthermore the very small amount of ascorbic acid necessary both for the prevention and cure of scurvy, fits well with this hypothesis.

When more vitamin C is supplied to the erythron, the picture does not change, except in degree. The reticulocyte levels rose promptly and sharply and the bone marrow, although similar to that prior to treatment, was the scene of intense activity.

It is possible that the pathogenesis of the anaemia of scurvy may not be so simple. It is possible that ascorbic acid may act indirectly on the erythron by regulating certain enzyme reactions in the final build up of the constituents of the red blood cell. The prompt and exceedingly rapid response to ascorbic acid, without any other factor being necessary before, during or after such treatment, furnishes facts sufficient to convince the unprejudiced that vitamin C is essential for normal erythropoeisis.

SUMMARY OF THE ANAEMIA OF SCURVY.

1. It appears from this study that anaemia is of high incidence in adult scurvy.

2. This anaemia can be very severe and responds specifically to pure synthetic ascorbic acid and no other factor.

3. Vitamin C deficiency appears to produce its anaemia in adults after a prolonged deficiency, well after the signs of clinical scurvy are manifest.

4. It is suggested that this anaemia results from a pure deficiency, i.e. no other concomitant deficiency is necessary for its production.

5. The diagnosis of the anaemia of scurvy is recognised by its morphology and associated features:-

(a) Clinical signs of scurvy.

(b) A normocytic, occasionally macrocytic normochromic anaemia with a peripheral blood smear in severe cases, that shows anisocytosis and "anisochromia", but rarely poikilocytosis. Platelets are in normal numbers and rarely nucleated red blood cells are seen.

(c) Reticulocytes may be present in increased numbers.

(d) Urobilinogenuria and raised faecal urobilinogen levels may be found if associated liver dysfunction and bleeding into the tissues is present.

(e) The serum iron levels and the iron binding capacity may be low.

(f) Often achlorhydria which may be histamine-fast, but with pepsin present in normal quantities is associated.

(g) The bone marrow although hypercellular in appearance shows decreased mitosis. Occasionally megaloblasts are seen.

(h) A rapid and complete response occurs with ascorbic acid therapy.

GENERAL SUMMARY.

Certain aspects of scurvy have been surveyed on the basis of the findings in thirty two adult males.

I. A brief outline historically showed that scurvy was a menace to man even, probably, in Biblical times. Attempts to combat this menace shaped the course of history itself.

2. That Cape Town , established as an antiscorbutic station 300 years ago, now provides one of the largest series of adult scurvy of recent times is not only paradoxical but disturbing. It is disturbing because scurvy appears to be on the increase as judged by the hospital admissions. The suggested reasons for this are an increasing exodus of the unskilled rural Bantu to the towns and a rising cost of living. The Bantu customs and superstitions are still the same,in the rural areas, as those encountered by anti-scurvy campaigners of 50 years ago. His health,still balancing precariously on a very low vitamin C intake, is preserved in the kraals. He does not like fresh vegetables or fruit, whether they are plentiful or not. This balance may be tipped against him when increased demand for, or increased loss of the vitamin e,g. in the sweat, occurs. This may explain the greatest frequency of admissions occurring in the late summer .

3. Apart from this, it was felt that it was unnecessary when considering the etiology of these cases, to postulate other predisposing factors, such as infection or failure of absorption despite a high incidence of achlorhydria which persisted in some, even after the anaemia had been corrected. Evidence exists that

before the vitamin leaves the Bantu's pot, it would have been thoroughly destroyed by his haphazard methods of cooking. In analysing the diet to which he is accustomed, it is felt that even to postulate this is somewhat unnecessary. The racial incidence was in support of this. It was indeed striking that from one of the compounds, predominately of Xosa population, all patients were Nyasas, who did not partake of this Kaffir Beer. In order then, to combat the problem of this, a near absolute vitamin C deficient diet, it is suggested , as has been over the last fifty years, that until the Bantu learns better dietetic habits, this beverage of low alcoholic content but containing vitamin C, should be encouraged in the compounds. Encouraging is the sight of the work being done since the recent establishment of Health Centres in the heart of the Native reserves. It is encouraging in that the cause of this ignorance is being eradicated at its source.

4. The clinical syndrome of adult scurvy is fashioned on skin manifestations, gum manifestations, and deeper haemorrhagic manifestations. They appear in that order.

(a). The first clinical sign is follicular hyperkeratosis, followed later by perifollicular petechiae. Only at this stage does any interference with normal wound healing occur. The practice of giving ascorbic acid routinely pre-operatively, in the absence of follicular hyperkeratosis is, then, both unnecessary and uneconomical.

(b). Gum manifestations are not reliable. The edentulous show no such features whereas those with teeth may have no changes, or changes so localised that a diligent search is necessary. On the other hand the gingival hypertrophy may be so massive as to cover the teeth, or appear as large granulomatous excresences extending onto the palate. Bleeding, pain and halitosis may or may not accompany this hypertrophy.

(c). Deeper haemorrhagic manifestations are the haematomata and ecchymoses characteristic of the disease. These present all the cardinal features of inflammation with distal oedema or neighbourhood joint effusions. Bleeding from elsewhere, e.g. mucous membranes of the nose, gut, etc., or into serous cavities, may occur.

Another accompanying feature frequently found is anaemia which, by giving rise to many secondary effects, causes the difficulty in assessing the specificity of other scorbutic features, such as low blood pressure, pyrexia, and water retention Concerning the last-mentioned, it was suggested that further study may relate this to the reported diminished glomerular filtration which occurs in severe scurvy, probably to attempt to conserve all available vitamin C.

5. The type of diet on which scurvy occurs, may lead to concomitant liver dysfunction and other avitaminoses. There

is no evidence to suggest that the constituents of the diet condition this deficiency as a high carbohydrate diet may condition the appearance of the vitamin B deficiency.

6. The minimal daily requirement of vitamin C is in the region of or somewhat below 10 mg. That vitamin C is stored in the body and that these stores continue to supply the body needs for a long time, in the absence of exogenous supply, has been shown. These two facts adequately explain many of the unsolved problems of scurvy of earlier days. i.e.

(a). The long incubation period.

(b). The rarity of scurvy amongst populations all existing on a diet very low in vitamin C content.

(c). The clinical response that occurs merely on putting the patient to bed, and the adequate response to a mixed hospital diet only.

7. A subclinical scorbutic state giving rise to ill health has yet to be proved. Until then, this concept should be dropped as it merely leads to confusion and an unnecessary waste of synthetic vitamin C.

This state is only diagnosable on technical or chemical tests, all of which, with the possible exception of the white cell-platelet layer estimation are, totally unreliable.

The only reasonable conclusion is that an "unsaturated" person is a potential scorbutic.

8. This "unsaturated" state has helped to confuse

the issue of ascorbic acid and erythropoeisis. This confusion is well reflected in the recent reviews of the subject and in the standard text-books.

After a detailed critical analysis of the literature it was felt that the following points caused this confusion:-

(a). Ascorbic acid has no proven effect against any condition other than scurvy.

(b). Both anaemia and scurvy may result from dietary deficiencies and to prove that one is the result of the other needs, above all, a very strict dietary control. The simplest and yet most accurate controlled diet would be that on which the disease developed.

(c). Anaemia due to vitamin C lack is due to chronic vitamin C lack in the form of clinically obvious scurvy and therefore this does not mean merely the chemical state of "unsaturation".

(d). Influences exerted by haemorrhage, other deficiencies and by diminished metabolic requirement (as occurs with bed-rest) should first be eradicated before the effect of pure synthetic ascorbic acid on erythropoeisis is assessed.

Observing these principles, a prompt and complete response followed, in ten consecutive cases in this series, the addition of pure ascorbic acid only, without any other factor, such as adequate amino acid intake, iron, extrinsic

factor, or other vitamin, being necessary before, during, or after in addition, whether they were deficient in the diet or not. Folic acid, vitamin B_{12}, iron and/or vitamin B complex failed to have any influence on the bone marrow or blood picture.

It is felt, then, that in the adult it has been proved now, beyond doubt, that vitamin C is essential for normal blood formation.

9. An attempt was made to determine the nature of the anaemia.

(a). High pigment excretion levels, suggestive of a haemolytic process were shown to be due to the extra-vascular haemolysis in the haematomata with concomitant liver dysfunction.

(b). The failure, of others, to produce anaemia by venesection in a mild scorbutic was strong evidence against the anaemia being due merely to acute blood loss. The morphology and the failure of any response to iron helped to exclude chronic blood loss as the mechanism.

(c). The dyshaemopoeisis was not thought to be toxic in origin although a similarity to the anaemia of infection was noticed. This similarity lay in the low plasma iron and iron binding capacity. It was felt that the intramuscular haematoma was responsible for these effects.

Morphologically the anaemia is normochromic and normocytic

but occasionally macrocytic in the more severe cases. The normal D:T ratio and the infrequency of target cells make liver disease unlikely as the cause of this macrocytosis. Variation of the size and haemoglobin content of the red cell becomes more noticeable as the anaemia progresses. Variation in the shape was less frequently seen. Reticulocytes may or may not be present in increased numbers in the severer cases. Once bed-rest is instituted this feature is constantly seen. Nucleated red cells rarely are seen in the peripheral smear.

Platelets occur in normal numbers.

The bone marrow, although often hypercellular in appearance, shows decreased mitosis. Occasionally megaloblasts are seen.

A histamine-fast achlorhydria with normal pepsin activity and a leucopaenia may combine with the above features to show a close similarity to other deficiency dyshaemopoeitic anaemias of the megaloblastic type.

Finally the proof of the diagnosis lies in a rapid and complete response to ascorbic acid therapy.

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APPENDIX I.

DETAILS OF METHODS.

A.

TESTS USED FOR ASSESSMENT OF LIVER FUNCTION.

1. Palpation of the Liver.
2. Icteric Index.
3. Serum van den Bergh Reaction.
4. Serum bilirubin.
5. Serum Proteins:
    - (a) Total
    - (b) Serum Albumen
    - (c) Serum Globulin
    - (d) Albumen Globulin ratio
6. Thymol Turbidity of the serum
7. Thymol Flocculation of the serum
8. Serum Colloidal Gold
9. Serum Alkaline Phosphatase
10. Serum Cholesterol
11. Urine for Bile pigments
12. Urinary Urobilin
13. Urinary Urobilinogen
14. Faecal Urobilinogen

DETAILS OF METHODS.

1. Palpation of the Liver.

This was carried out in the course of the physical examination. Assessment of enlargement was measured as centimetres below the costal margin in the plane of the ninth costal cartilage provided the upper level of liver dullness on percussion was at the normal level. At the same time in the personal series liver dullness was carefully traced out on percussion to confirm enlargement and at the same time to ensure that a decrease in the size of the liver was not present.

Where palpable the liver edge was noted as whether sharp or round and whether it followed its normal oblique outline across the epigastrium or whether the enlargement was more obvious in the left lobe as advised by Himsworth (1948). Furthermore the surface was assessed as hard, firm or soft, smooth or nodular. Finally tenderness, if present, was noted.

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10 ml. of plain blood was drawn into a clean dry test tube, allowed to clot and the extracted serum used for the following tests. Haemolysis was as far as possible avoided.

2. Icteric Index.

A Klett-Summersen photo-electric colorimeter was standardised with 1 in 10,000 potassium dichromate solution. The diluted plasma was read against a blank of distilled water. The normal range was taken to be 4 to 7 units.

3. Serum van den Bergh's Test - Direct Reaction.

1 ml. Serum was treated with 0.5 ml. freshly prepared diazo reagent. If no colour change was noted within half an hour the reaction was recorded as negative. If however the red colour developed within thirty seconds, the reaction was recorded as prompt positive.

4. Serum Bilirubin (Indirect quantitative van den Bergh).

The method of MALLOY AND EVELYN (1937) was used for this determination. The result was expressed as negative, slight trace or in mg. per 100 ml. if the amount was 0.5 mg. per 100 ml. or more. The upper limit of normal in this laboratory using this method is 1.2 mg. per 100 ml.

5. The total serum proteins including the albumen and globulin fractions.

These were estimated using WEICHSELBAUM's method (WOLFSON et al 1948), the result being expressed as G. per 100 ml.

The normal range of Total Proteins using this method in this laboratory was from 6.0 G. to 8.5 G per 100 ml., with the albumen fraction from 3.5 G to 6.7 G per 100 ml. and globulin 1.2 G. to 3.0 G per 100 ml.

From the latter the albumen-globulin ratio was calculated. The normal ratio varies between 4:1 and 1.2:1.

6. Thymol Turbidity reaction.

This was performed according to the method of

MACLAGAN [1944(b)] using a Klett - Summersen photo-electric colorimeter. The result was expressed in units.

The upper limit of normal for this method in this laboratory is 4 units.

7. Serum Thymol Flocculation reaction.

The above tubes were then left in the rack and read twenty-four hours later. The result was expressed as an index from 0 with no flocculation to 4 where complete flocculation with no supernatant turbidity.

The normal level here is regarded as 0.

8. Serum Colloidal Gold reaction.

MACLAGAN's method [1944(a)] was followed. The mixed gold solution and diluted serum was allowed to stand and the result expressed as an index from 0 where there was no precipitation i.e. the red colour persisting, to 5 where complete precipitation had occurred.

The upper limit of normal in this laboratory is 0.

9. Serum Alkaline Phosphatase.

This was done according to the method of SHINOWARA et al (1942). The result was expressed in units per 100 ml. serum.

The normal limits as done by this method vary from 2.2 Bodansky units per 100 ml. to 8.6 units per 100 ml. for adult males.

10. Serum Cholesterol (Total).

Estimation of serum cholesterol was performed according to the method of ANDES (1935).

The result was expressed in mgm. per 100 ml. serum and the normal limits with this method vary from 120 to 220 mg. per 100 ml.

11. Urine for bile pigment.

(a) Colour.

A test tube of urine was shaken and the froth examined. A greenish yellow colour was expressed as positive i.e. bile pigments being present [ZWARENSTEIN and PALLEY (1946)].

(b) Fouchet's Test.

Equal quantities of acidified urine and 10% barium chloride solution were mixed and filtered. The precipitate on the filter paper was dried and one drop of Fouchet's reagent was added. A green or blue colour was regarded as positive in that it denoted the presence of bilirubin. At the same time the colour of the surrounding precipitate was noted with regard to the presence of urobilin as noted by HARRISON (1947).

12. Urine for Urobilin pigment.

The filtrate of the above Fouchet's test was examined by a spectroscope and if no characteristic absorption band at the junction of the green and blue was

present, four drops of Tincture of Iodine were added. The mixture was shaken and examined again spectroscopically. If no band appeared the result was expressed as urobilin not present.

13. Urinary Urobilinogen.

The quantitative urinary urobilinogen estimation was considered necessary as most variable results were noted in the previous cases using the spectroscopic or qualitative estimation of urobilinuria. STEIGMANN and DYNIEWICZ (1943) in comparing various methods on over a thousand urines found that Watson's method [1936, 1937, 1944(a), 1944(b)] was more accurate than SPARKMAN's (1939). Furthermore, as the methods adopted by other workers on Scurvy were those of Watson, Watson's method was adopted where the colour produced by the addition of Ehrlich's Aldehyde reagent to urine was estimated as reflecting the urobilinogen content. Due to the warnings of WILSON and DAVIDSON (1949), reagents of high analytical standard were used throughout to prevent any possibility of false positives interfering with the results obtained. Likewise care was taken, as shown by KELLY et al (1946) and WILSON and DAVIDSON (1949) that the sodium acetate in the direct test was added as soon as possible after the Ehrlich's reagent so that false reactions from substances other than urobilinogen were largely eliminated.

Twenty-four hour urine collections were preserved with sodium carbonate under a layer of petroleum ether in dark

containers. After thorough mixing 50 ml. had 25 ml. of freshly prepared 20% Ferrous Sulphate and 25 ml. of 10% NaOH added to it. The mixture was allowed to stand in the dark for one to three hours, depending on the presence or absence of the urobilin band spectroscopically in the filtrate.

The less time consuming and less expensive direct method was then performed daily. Equal quantities (2 ml.) of urobilin-free filtrate and Ehrlich's aldehyde reagent were placed in a test tube and 6 ml. saturated sodium acetate were added immediately afterwards. The colour produced was read against a blank using 6N HCl instead of Ehrlich's reagent in a Klett-Summersen photo-electric colorimeter, at 560 mu, standardised with Terwen's solution, as advised by MACLAGAN (1946). This solution is equivalent to .387 mg. crystalline mesobilirubinogen per 100 ml. The final calculation was facilitated by these quantities and the result expressed as Ehrlich units per day. The final coloured solution was shaken with chloroform and if it was not chloroform soluble the result was discarded and the petroleum ether method used.

The petroleum ether method was performed daily if the direct method was chloroform insoluble, otherwise it was performed every second or third day to check the results obtained by the quicker direct method.

The filtrate was shaken vigorously through petroleum ether three times, and then extracted from the petroleum ether with Ehrlich's reagent. Sodium acetate was

not added immediately. Extractions were repeated until no colour remained. The final coloured solution was then read as before in the photo-electric colorimeter using a blank of sodium acetate solution and Ehrlich's reagent only, as advised by WATSON (1937). The final calculated result was expressed as mg. urobilinogen per day.

14. Faecal Urobilinogen.

Faeces were collected in weighed waxed containers and tested immediately. The container was weighed again. A portion (1.5 G) was removed after thorough mixing; the container made airtight and stored in a refrigerator for Watson's four day estimation [SCHWARTZ, SBOROV and WATSON (1944)]

Daily estimations on the 1.5 G portion were estimated as advised by MACLAGAN (1946). The faeces were ground up into an emulsion with 9 ml. distilled water and stored for an hour to three hours in the dark with a ferrous hydroxide mixture as used for the urine. A 2 ml. portion of the urobilin-free filtrate was treated with Ehrlich's reagent (2 ml.) with 6 ml. sodium acetate added immediately as in the urine test. A suitable dilution (usually up to 100 ml.) was made of the final coloured solution. This was then read in the photo-electric colorimeter in the usual way. The final calculated result was expressed as advised by MACLAGAN (1946) as mg. per 100 G.

The weighed four-day collection was thoroughly

mixed. An emulsion was made with a 10 G. portion. This was treated with ferrous hydroxide in the usual way and petroleum ether extraction performed as for urine. The final calculated result was expressed as advised by WATSON (1937) SCHWARTZ, SBOROV and WATSON (1944) as mg. per day.

DISCUSSION OF METHODS.

The normal limits and methods of tests 3 to 9 were obtained from the department of Chemical Pathology, University of Cape Town. I am indebted to Professor Linder who performed these tests and allowed me to use his results.

The Icteric index was instituted later in the series as the serum often appeared yellow to the naked eye without any bilirubinaemia. The icteric indices were elevated. WATSON (1937) describes a similar state of affairs in a case of a haematoma of the kidney which he attributes to "haematin icterus". Unfortunately the presence of haematin could not be ascertained with the methods available.

With regard to the Urobilinogen studies, photo-electric colorimetric methods were substituted for visual colorimetry which is subject to the wide range of variation in reading of results.

The photo-electric colorimeter was standardised anew every month with freshly prepared Terwen's solution.

The probable accuracy of the direct rapid method was studied by WILSON and DAVIDSON (1949) in urines. The substances liable to give false reactions are indole, the precursors of two indole pigments - indirubin and urorosein - and porphobilinogen. Urorosein and porphobilinogen-aldehyde are insoluble in chloroform. If the hydrochloric acid

employed in the reagent is pure and free from oxidising agents and the reagent is used in small amounts or if the acid is immediately neutralised with sodium acetate - a procedure considered important by KELLY et al (1946) as well - the reddish pink colour developing is unlikely to be due to any substance other than urobilinogen.

This advice was rigidly followed in this series. Additional accuracy was obtained by using 6N HCl as a blank instead of Ehrlich's reagent. The same hydrochloric acid (C.P.) was used in both, consequently any false reaction due to urorosein would be seen in the blank.

The rapid addition of acetate and converting the hydrochloric acid to Acetic Acid [KELLY et al (1946), WILSON and DAVIDSON (1949)] inhibits colour reactions due to indole and possibly indirubin [WATSON (1936)].

Fortunately as WATSON (1937) and MACLAGAN (1946) point out the increase of these non-urobilinogen reacting substances is roughly proportional to the increase in urobilinogen, probably having similar significance.

To ensure greater accuracy however, the possibility of the remaining false reactor-porphobilinogen made chloroform extraction necessary. If the pimk colour was not completely soluble the more time consuming petroleum ether extraction was performed daily instead of only as a check every second or third day.

Other possible inaccuracies due to evaporation were carefully watched. This applied more to faeces. WATSON (1937) states that there is a 7% loss over the four days if the faeces are kept in closed containers at 4°C. The considerable day to day fluctuations when the patient was not receiving a constant diet and the variability in frequency of bowel movements in different individuals makes imperative the determination of the average amount excreted over several days [WATSON (1936)]. To ensure daily bowel action magnesium sulphate was given every morning. STEIGMANN and DYNIEWICZ (1943) found that this did not interfere with the urobilinogen content. These authors together with MACLAGAN (1946) have shown that random specimens expressed as mg. per 100 G. were of equal value as the four day collection expressed as mg. per day. The similarity in quantity depends on a daily stool weight of 100 G. MACLAGAN (1946) mentions that the differences are only considerable in cases with low or normal values. WATSON et al (1944) comment that from a clinical standpoint the information gained is equivalent to that obtained with the exact quantitative method but for more precise results the petroleum ether method is preferable.

The normal values of other investigators using WATSON's Ehrlich reagent (0.7 G. paradimethylaminobenzaldehyde, 150 ml. HCl 100 ml. distilled water) are compared with values obtained on thirty estimations on normal people or patients in whom no increase in urobilinogen was expected. Three of

the latter were Bantu males on the same diet of mealie meal porridge as given to the scorbutics. The estimations were performed using the same apparatus and chemicals as used in the scorbutics.

AUTHOR	DIRECT METHOD		PETROLEUM ETHER METHOD	
	URINE	STOOL	URINE	STOOL
Steigmann and Dyniewicz (1943)		15 to 175mg./100G (85% between 30 & 120)	0.2 to 3mg./day (80% between 0.4 & 1.7)	20 to 200mg./day (74% between 40 & 120)
Maclagan (1946)		30 to 220mg./100G. (22 to 121mg./day over 4 day period)		
Watson (1937) (1944)	1.12 to 4.9 units per 24 hrs.	120 to 276mg./100G	0 to 4mg./day (Usually 0.3 to 1.7)	40 to 280mg/day (Usually 100 to 250mg./day
This Series	1.4 to 5.2 units per day	20 to 180mg./100G. (78% between 40 & 130)	0.15 to 1.8mg./day (70% under 1.0mg./day)	12 to 165 mg./day (83% between 25 and 100)

This comparison serves to illustrate the probable accuracy of the methods used and serves as a standard for the

results obtained in Scurvy. It will be seen as did MACLAGAN (1946) that somewhat lower figures were obtained with the petroleum ether extraction method for four day stool collections. This may be explained by the fact that the average weight of daily faeces for the thirty investigations was 54 G. per day. It was noticeable that on the three Bantus on mealie meal porridge the daily faecal amounts were lower than any other group.

Mention may be made that STEIGMANN and DYNIEWICZ (1943) draw attention to the importance of noting the faecal-urinary urobilinogen ratio in assessing liver function. Normally this is 100 : 1 to 40 : 1. A low ratio demonstrates that the liver is not able to remove even small amounts from the blood.

B.

METHODS USED FOR ASSESSING VITAMIN C SUBNUTRITION.

1. Capillary fragility test.
2. Intradermal Dye test.
3. Plasma Ascorbic acid.
4. Urinary Ascorbic acid.
5. "Saturation" tests.
6. Ascorbic acid "tolerance" test.
7. Ascorbic acid content of White cell platelet layer.

DETAILS OF METHODS.

The haemorrhagic tendency that occurs in Scurvy and the fact that ascorbic acid is a powerful reducer of the dye 2:6 dichlorophenolindophenol has led to the development of numerous tests to detect vitamin C subnutrition.

1. Capillary fragility test.

DALLDORF (1931) showed in guinea pigs that the haemorrhagic diathesis in experimental scurvy develops earlier than any other known sign of the disease and that it persists in some degree throughout. He showed this by applying negative pressure cuffs on the belly wall. It is of interest to mention that occasionally haematomata occurred instead of petechiae and this tendency increased as the disease progressed.

Various methods have been tried in an attempt to demonstrate this in man by applying positive or negative pressure to the arm or elsewhere. All have different names e.g. RUMPEL-LEEDE, HESS (1920), GOTHLIN (1937) test etc., with varying standards. All measure the fragility or resistance of the capillary walls by counting the number of petechiae so produced in a given area.

LIEBMAN, WORTIS and WORTIS (1938) in assessing the various tests mentioned above advised Wright's modification of the Rumpel-Leede test [WRIGHT and LILIENFIELD (1936)]. This was the test used in this series. A blood pressure cuff was applied to the arm. A circle one inch in diameter was

drawn distal to the cuff on the antecubital fossa; all artefacts were marked to ensure that no previously existing marks would be counted as petechiae. If possible an area free of such marks was selected. The cuff was then pumped up so that the pressure lay between systolic and diastolic blood pressures and it was maintained there for fifteen minutes. Five minutes later petechiae if present were counted in this circle. The normal with this method is less than ten petechiae, whilst between ten and twenty petechiae are regarded as borderline; more than twenty indicate a positive test.

2. Intradermal Dye test.

This test depends on the ability of ascorbic acid to reduce 2.6 dichlorophenolindophenol and was developed by ROTTER (1937). He noticed that when small quantities of the dye were injected into the soles of guinea pigs decolourisation occurred more rapidly in normals than in scorbutic animals whereas methylene blue similarly injected remained unchanged. From this he concluded that the mechanism was reduction and not resorption of the dye.

0.01 ml. of the dye solution (of strength 2 mg. in 4.9 ml. [WILKINSON and PORTNOY (1938)]) is injected intradermally in four places on the forearm. Normally the blue spots are decolourized within ten minutes. This test was abandoned in this series.

3. Plasma ascorbic acid.

This investigation with the patient fasting is

performed on plasma soon after taking as possible. Several methods are available including micro methods [FARMER and ABT (1936)]. Each method is the same in principle i.e. precipitation of protein and determination of the degree of reduction of 2:6 dichlorophenolindophenol by the protein free filtrate. This reduction can be measured by titration or on a photo-electric colorimeter. Haemolysis must be avoided. The method of KING (1946) was used in this series. The normal figures vary considerably. 0.5 to 1.0 mg. per 100 ml. is regarded by EDDY (1941) as borderline and above 1.0 mg. per 100 ml. as normal whereas PORTNOY and WILKINSON (1938) regard any level above 0.6 mg. per 100 ml. as normal.

4. Urinary Ascorbic Acid.

Precautions have to be taken in this test that no marked loss of ascorbic acid occurs during the twenty-four hour collection. Consequently it is advised [ABBASSY et al (1935)] that the urine be kept in stoppered dark bottles and glacial acetic acid be added so that the final concentration of glacial acetic acid is approximately 10%. This may be achieved by adding one part glacial acetic acid to nine parts of urine as each voiding is added to the bottle [KING (1946)].

For the actual determination 0.1 ml. of 2:6 dichlorophenolindophenol dye, which has not been prepared longer than two weeks, which has been stored in the cold and is of a known concentration standardised against pure ascorbic acid, is used. The dye is acidified with one drop of glacial acetic

acid and titrated rapidly with urine according to the method of KING (1946). The volume of the twenty-four hour urine is measured and in the calculation allowance is made for the amount of acetic acid added.

An excretion above 20 mgm. ascorbic acid a day is regarded as normal.

Other methods, one including the use of a photo-electric colorimeter, have been suggested [EVELYN et al (1938), ROE and HALL (1939)]. These methods still depend on the rapid reduction of the indophenol dye.

5. "Saturation" tests.

The rationale of this test is based on the assumption that when a test dose is given, the tissues and kidneys compete for the available blood ascorbic acid. When the tissues are satisfied more and more of the test dose is poured out into the urine.

Many different methods unfortunately are in use. The route of administration varies considerably some preferring the oral route [ABBASSY et al (1935)] and others the intravenous route [WRIGHT et al (1936), PORTNOY and WILKINSON (1938)]. The amount of the test dose varies considerably and the criteria for the end point varies with each individual worker.

As the uncertainty of adequate intestinal absorption existed, the intravenous method as advised by

WRIGHT et al (1936)(1939) was used in this series.

The patient was told to empty his bladder and 1000 mg. ascorbic acid was given by slow intravenous injection. The slowness of the injection, as pointed out by BROCK (1942), FOX et al (1940) and others, is to prevent a sudden rise in the plasma ascorbic acid above the renal threshold. All urine voided within five hours of the injection is analysed for its ascorbic acid content. The patient was said to be "saturated" when he excreted more than 40% of the injected dose (i.e. 400mg.) in the five hours following injection.

Most authors consider that a healthy person should be saturated within two days.

6. Ascorbic acid "tolerance" test.

This test embodies the same principles as the "saturation" test. KASTLIN et al (1940) recommend 500 mg. ascorbic acid intravenously. The plasma ascorbic acid levels are determined five minutes, one hour, two hours, three hours, four hours and five hours thereafter. If the tissues are "saturated" they will not take up ascorbic acid rapidly, consequently the rise in plasma ascorbic acid will be rapid and high and it will fall slowly. In an "unsaturated" state the tissues will take up the ascorbic acid greedily, consequently the blood level will not rise so high and will soon fall to normal levels.

Unfortunately there are many variations in route of administration and in dosage [PORTNOY and WILKINSON (1938)].

No such test was performed in this series.

7. Ascorbic acid content of White cell - Platelet layer.

This test was developed by BUTLER and CUSHMAN (1940) but the method is both tedious and difficult and not readily adaptable for routine clinical use. Unfortunately these reasons prohibited its performance in this series. The test was developed due to the conflicting evidence regarding whole blood analytical procedures.

In a special centrifuge tube with a constricted waist blood is centrifuged and the white cell-platelet layer removed and the degree of the reduction of 2:6 dichlorophenol-indophenol is measured. The ascorbic acid level here is between twenty and forty times the concentration in the plasma. A level below 2 mgm. per 100 G. on repeated estimations indicates severe depletion of Vitamin C and supports the diagnosis of scurvy [PETERS et al (1948)].

A full discussion on the value of these tests appears earlier in the main body of this thesis.

C.

OTHER TESTS USED BECAUSE OF THE BLEEDING TENDENCY.

1. Bleeding time.

2. Coagulation time.

3. Blood Platelet counts.

4. Prothrombin time.

DETAILS OF METHODS.

1. Bleeding time.

Duke's method of estimation was adopted in this series [WINTROBE (1946)]. More than five minutes was regarded as prolonged.

2. Coagulation time.

The method of Lee and White was adopted where the normal range extends from six to fifteen minutes [WINTROBE (1946)]. Control estimations were performed at the same time.

3. Blood platelet counts.

Two methods were used as recommended by WINTROBE (1946): (a) The rough and simple method of noting the number of platelets as seen on a dry stained smear for a differential count was used in each case; (b) The direct method of counting platelets by using a clean dry red cell counting pipette was used for the more accurate estimation in the personal series. Diluting fluid (containing brilliant cresyl blue) was drawn up to the 0.5 mm. mark, the finger was cleaned, dried and punctured. After wiping the first drop away, blood was drawn up so that the column of blood and diluting fluid in the pipette was at the 1.0 mark. Diluting fluid was then drawn up to the 101 mark. The pipette was thoroughly shaken. Platelets were then counted on the red cell counting slide, adopting the usual precautions.

Values between 200,000 and 500,000 per cu. mm. were regarded as normal [WINTROBE (1946), WHITBY and BRITTON (1942)].

4. Prothrombin times.

On seven of the thirteen cases in the personal series this estimation was performed. The method of QUICK (1935) was used. Control determinations were performed at the same time. Unfortunately the middle six cases could not have this estimation performed as no satisfactory thrombokinase was available at that time.

D.

METHODS USED IN THE HAEMATOLOGICAL INVESTIGATION.

The following procedures were performed:-

1. Red cell count.
2. Haemoglobin estimation.
3. Packed cell volume.
4. White cell count.
5. Differential White cell count.
6. Reticulocyte count.
7. Erythrocyte sedimentation rate.
8. Measurement of the Red cell Diameters.
9. Hypotomic saline fragility test.
10. Fractional test meal.
11. Urinary urobilinogen.
12. Faecal urobilinogen.
13. Serum van den Bergh direct and quantitative indirect reactions.
14. Serum Proteins.
15. Liver Function tests.
16. Plasma Iron determinations.
17. Determination of Iron binding capacity of Plasma.
18. Bone marrow counts.

DETAILS OF METHODS.

1. Red cell counts.

Oxalated venous blood was used and the same "Spencer" counting slide was used throughout. The count was expressed as millions per cu. mm.

2. Haemoglobin estimation.

This was performed on a Dare haemoglobinometer. Later on in the series the accuracy of this haemoglobinometer was checked against a M.R.C. Grey wedge photometer. Although there was a less wide range with the latter, the results were comparable. The results were expressed in G. per 100 ml.

3. Packed cell volumes.

The haematocrit (P.C.V.) was measured according to the method of WINTROBE (1946) on oxalated venous blood, using Wintrobe tubes. Duplicate estimations were made and read to the nearest 0.5 mm. and expressed as a percentage.

4. White cell count.

This estimation was performed on oxalated venous blood using a Spencer counting chamber. The result was expressed as the number per cu. mm.

5. Differential White cell count.

Differential counts were performed on Leishman - stained films of capillary blood. Two hundred cells were counted and the result expressed as a percentage of each type of

White blood cell present.

6. Reticulocyte Counts.

Four drops of 1% aqueous solution of citrated brilliant cresyl blue was added to a small test tube. The finger was punctured and an equal quantity of blood was added to the test tube. This was thoroughly mixed and incubated at 37°C. for half an hour when smears were made. The number of reticulocytes per 1000 red cells was counted and the result expressed as a percentage.

7. Erythrocyte Sedimentation Rate.

The method of Westergren was used in each case. Into a syringe containing 0.4 ml. of 3.8% sodium citrate 1.6 ml. venous blood were added. Westergren tubes were filled and the level, taken after exactly one hour, was read as millimetres (West.) per hour.

8. The measurement of the Red Cell Diameters.

Two methods were used.

(a) Price Jones method.

Thin smears of peripheral blood were stained with Leishman's stain and counter-stained with 1% Eosin. A Standardized graduated slide (1 u = .01 mm.) was projected on to a ground glass screen of the camera of the microscope. Using a protractor the magnification was calculated. It was 1,750.

The smears were then projected on to the ground glass screen and measured by the protractor as suggested by WHITBY and BRITTON (1942). The sizes of the rings on the

protractor were then checked again after the manoeuvre. Five hundred cells were measured and graphed in the usual way.

(b) Haden-Hausser Erythrocytometer.

Using the diffraction method of PIJPER (1947) the same slides were measured. It was used for follow up studies.

The mean cell diameter was expressed in u in both methods but where the diffraction method was used the result was expressed as "x u (diffraction method)."

9. Hypotomic Saline Fragility Test.

The method adopted here was that advised by WHITBY and BRITTON (1942). The results were expressed as increased or decreased resistance to saline, or normal.

10. Fractional Test Meal.

After fasting overnight, a Ryle stomach tube was passed and the gastric contents aspirated. Caffeine Citrate (200 mg. in 300 ml. of water) was given by mouth and specimens withdrawn from the stomach at fifteen minute intervals for three hours. The sixth specimen was tested for acid and if none was present histamine acid phosphate 0.5 mg. was injected subcutaneously.

The free acid was titrated in the standard way with N/10 NaOH using Topfer's reagent as indicator. The results were expressed in the usual units i.e. ccs. of N/10 NaOH required to neutralize the acid in 100 cc. of gastric juice. The presence

of bile, blood and mucus was noted. These titrations were performed by Professor Linder to whom I am indebted for the results. The presence of pepsin was determined by Dr. O. Budtz Olsen using the Congo-Red fibrin method.

11. Urinary Urobilin and Urobilinogen.
12. Faecal Urobilinogen.
13. Direct and Indirect van den Bergh.
14. Serum Proteins.
15. Liver Function Tests.

The methods used have been described in the previous section (A)

16. Plasma Iron Determination.

These determinations were carried out by Dr. O. Budtz Olsen using his method [BUDTZ-OLSEN 1950(a)], the usual care being taken re the scrupulously clean glassware necessary in this determination. The results were expressed in micrograms (uG) per 100 ml.

17. Iron Binding Capacity determination.

Using the same technique the plasma iron was determined after an intravenous injection of iron [BUDTZ-OLSEN (1950)(a)].

18. Bone Marrow Counts.

Specimens of bone marrow were obtained by gentle suction through the needle inserted into the Iliac bone [RUBENSTEIN (1948)]. Smears were made immediately and stained with Leishman's stain. One thousand nucleated cells were

counted according to the classification of DACIE and WHITE (1949) for red cell precursors and OSGOOD et al (1948) for white cell precursors.

At the same time the degree of cellularity and mitosis was noted.

This examination was repeated after ascorbic acid therapy.

DISCUSSION OF TECHNICAL METHODS.

1. The Normal Cases.

A series of normals (see Table) including doctors, nurses, Bantu orderlies and patients suffering from conditions that are not supposed to affect the blood, demonstrate:-

(a) The probable accuracy of the red cell counts, haemoglobin estimations, packed cell volume estimations, reticulocyte counts and mean cell diameter (diffraction method) measurements in the series of cases of Scurvy here presented, as the normal cases were investigated at the same times, with identical techniques, solutions and apparatus.

Furthermore this probable accuracy is reflected in the constancy of repeated estimations every other day (as shown in the graphs previously) to obtain a base line before treatment was started.

(b) That the figures obtained from these normals compare favourably with normal figures elsewhere in the World.

(c) That no difference appears to exist between the normals for Bantu males than for males of other races.

(d) A standard for the anaemia associated with Scurvy.

Table B.

NORMAL FEMALES

Case No.	Age Yrs.	R.B.C. mill./ cu.mm.	HB G%	P.C.V. %	W.B.C. per cu.mm.	M.C.D. (Diffr.) u	Reticulo-cytes %
70	21	4.65	13.8	41	4800	7.0	0.6
71	25	4.86	14.4	44	7300	6.9	0.8
72	35	4.70	14.0	42	4900	6.9	0.7
73	24	4.60	14.0	41	5100	6.8	0.8
74	32	4.20	14.0	38	8800	7.6	0.8
75	41	4.63	14.6	43	5200	7.5	0.3
76	21	5.03	15.0	45	8800	7.0	0.3
77	21	4.40	15.0	40	5200	7.3	1.1
78	22	5.10	15.5	46	10100	7.3	0.4
79	56	4.32	13.0	40	5600	7.1	0.3
80	20	4.51	13.5	41	6700	7.5	1.2
81	55	4.90	13.8	40	5400	6.8	0.5
MEAN	31	4.66	14.2	42.6	6325	7.14	o.65
MIN.	20	4.20	13.0	38.0	4900	6.8	0.3
MAX.	56	5.10	15.5	46.0	10100	7.6	1.2

The first four patients in each table are Europeans.

The second four patients in each table are Coloureds.

The remaining patients in each table are Bantus.

Table A.

NORMAL MALES

Case No.	Age Yrs.	R.B.C. mill./ cu.mm.	HB G%	P.C.V. %	W.B.C. per cu.mm.	M.C.D. (Diffraction) u	Reticulocytes %
51	28	4.70	13.8	42	5600	7.5	1.2
52	26	5.31	16.0	48	9500	7.3	0.4
53	30	5.06	16.5	46	4800	6.8	0.3
54	26	5.41	16.0	50	8100	7.2	1.3
55	18	5.46	14.2	47	8100	6.8	0.3
56	36	4.76	14.0	43	7600	7.4	1.1
57	44	4.96	16.0	45	9200	7.0	1.8
58	26	4.51	15.0	40	9800	6.9	1.5
59	35	4.65	13.6	41	6700	7.4	0.9
60	30	4.84	14.0	43	8100	7.2	1.6
61	30	4.70	14.2	41	5000	7.2	1.6
62	18	4.80	12.6	41	7400	6.8	0.4
63	47	4.30	14.5	40	7750	7.6	0.5
64	31	5.31	16.0	48	7200	7.5	1.2
65	45	4.66	15.0	41	5200	6.9	0.3
66	28	5.82	17.0	54	8700	7.0	0.9
MEAN	31	4.95	14.9	44.4	7420	7.16	0.95
MIN.	18	4.30	12.6	40	4800	6.8	0.3
MAX.	47	5.82	17.0	54	9800	7.6	1.8

The first four patients in each table are Europeans.

The second four patients in each table are Coloureds.

The remaining patients in each table are Bantus.

2. Haematological Indices.

(a) Packed Cell Volume (P.C.V.)

By this is meant the volume of packed red cells after centrifuging at 3,000 r.p.m. for 30 minutes. The lower limit of normal for adult males was taken to be 40% for the purpose of this investigation [WINTROBE (1946)].

(b) Mean Corpuscular Volume (M.C.V.).

This was calculated from the red cell count and packed cell volume and the result expressed as cu. u. The normal limits for this investigation were taken to be 80 to 94 cu. u [WINTROBE (1934)].

(c) Mean Corpuscular Haemoglobin Concentration (M.C.H.C.)

This was calculated from the haemoglobin expressed in G.% and the packed cell volume. The accepted range was 32 to 38 expressed as a percentage [WINTROBE (1934)].

(d) Mean Corpuscular Haemoglobin (M.C.H.)

From the haemoglobin in G.% and the red cell count this was calculated and expressed in micromicrograms ($\gamma\gamma$). The normal variations are from 27 to 32 [WINTROBE (1934)].

(e) Mean Cell Diameter (M.C.D.).

This was expressed in u and calculated from 500 cell measurements. The normal range [PRICE JONES

et al (1935)] has been given as 6.654 to 7.686 u. The degree of anisocytosis was calculated from the standard deviation and coefficient of variation, while macrocytosis and microcytosis was calculated from the graphs as shown by WHITBY and BRITTON (1942).

(f) Mean Corpuscular Average Thickness (M.C.A.T.)

This was calculated in the usual way from the mean cell volume and the mean cell diameter. The normal range is said to be 1.7 to 2.5 u. [PRICE JONES et al (1935)].

(g) Diameter Thickness ratio.

Ratios below 2.4 indicate definite spherocytosis whilst a figure higher than 4.2 denotes a flat cell [WHITBY and BRITTON (1942)].

(h) The Leucocyte-Erythrocyte Bone Marrow Ratio (L:E ratio)

The total number of nucleated cells of the white blood cell series in the thousand cells counted was divided by the total number of nucleated cells of the red cell series. The result was expressed as a ratio. Normally in the adult the L:E ratio is 4 or 5:1 [WINTROBE (1946)]. White blood cell counts of peripheral blood were performed at the same time as the marrow punctures to ensure that no false impressions of red cell activity were gained e.g. Hyperplasia of the white cell portion could give a false impression of underactivity of erythropoeisis

and vice versa.

All things being equal, alteration of this ratio with the degree of cellularity was classed as hyperplasia, normal or hypoplasia. Furthermore, the type of early red cell led to the further addition of the adjective normoblastic or megaloblastic to hyperplasia.

Whilst performing the count the degree of mitosis in the red cell series was observed before and after treatment with ascorbic acid.

3. Factors that affect blood estimations.

WINTROBE (1946) considers these factors as important:

(a) Age.

This has been excluded by studying only adults.

(b) Sex.

Only males were studied.

(c) Diurnal variations.

An attempt was made to ensure that all investigations were performed at a fixed time each day.

(d) Exercise.

All patients except one were bed patients. The one exception was rested for at least an hour before the estimation was performed.

(e) Barometric Pressure.

All cases were studied in the same section of Groote Schuur Hospital, Cape Town.

(f) Dehydration.

This was not clinically detectable in any of the cases studied except in one whose packed cell volume was above normal. Reference will be made to this under his case history (Case 7 Appendix II).

(g) Climate, Temperature and Season.

All estimations were done with the patients warm in bed. No control was possible concerning the season. WINTROBE (1946) however does not feel that these factors have a significant effect.

4. Conclusions re Haematological Technical Methods.

(a) Reticulocyte Counts.

The probable inaccuracy of single estimations of reticulocyte counts was overcome by repeated estimations. In that way a base line was obtained and significant variations or responses to treatment were easily detectable. The normal upper limit was taken to be 2%.

(b) The Red cell count, Packed cell volume and Haemoglobin estimation.

It was felt that greater accuracy would be obtained if

packed cell volume estimations were repeatedly estimated, although it meant venipuncture and the loss of at least three ml. of blood every second or third day. BIGGS and MacMILLAN (1948) found a minimum error of 9.46% in red cell counts i.e. with an initial count of 2,000,000, subsequent counts must be under 1,400,000 or over 2,600,000 before a significant fall or rise can be claimed. Naturally the colour index, mean corpuscular volume and mean corpuscular haemoglobin reflect this inaccuracy. The minimum error for haemoglobin is, on the other hand, 5.2% whereas for the haematocrit it is only 0.5%. Consequently the M.C.H.C. has a greater degree of accuracy than other indices. Regarding the morphology it was considered that a detailed inspection of the peripheral smear for alterations in the size (anisocytosis) and shape (poikilocytosis), alterations or variations in the haemoglobin content (anisochromia) together with the mean cell diameter with its attendant indices as determined by the Price-Jones method was a far more accurate assessment than the indices mentioned above, with the possible exception of the mean corpuscular haemoglobin concentration.

E.

OTHER TESTS PERFORMED.

1. Blood Urea.

This was performed by Professor Linder using Archer and Robb's method [HARRISON (1947)]. The normal ranging from 20 to 40 mg. per 100 ml.

2. Serum Calcium.

This was also performed by Professor Linder using the oxalate-permanganate method [HARRISON (1947)].

A P P E N D I X II.

CASE HISTORIES
OF CASES FOLLOWED UP IN
PERSONAL SERIES.

C A S E : 1.

No. 137046. Bantu male (Nyasa) aged 33 years; admitted on 26th March, 1949; discharged on 10th May, 1949.

HISTORY:- For one month he has noticed an ulcer on his left ankle which is getting larger. It began as a small abrasion from chafing of his shoe. Clinical out patient diagnosis was squamous epithelioma but biopsy revealed chronic inflammation.

For the last three weeks he has had pain in the right elbow region and has difficulty in straightening the elbow. There is also pain in the right thigh which is worse on movement. Over the same duration his gums have bled on eating bread.

DIET:- Labourer; lives in a compound; has to provide his own food and cook it. Bread, mealie meal porridge, fish once a week and occasionally meat, for over five years.

EXAMINATION:- Pallor marked, bright mentally and co-operative. Painful limitation of movement of right elbow, some thickening in cubital fossa region, right thigh and left calf and tenderness. No large haematomata were obvious. Follicular hyperkeratosis present. "Spongy" gums, halitosis with septic teeth and pyorrhoea. Pyrexial, tachycardia with collapsing pulse B.P. 110/55 mms. Hg. Systolic murmurs over praecordium. Fundus oculi pale.

Stationary ulcer 1½ inches in diameter with

necrotic surface just anterior to and above left medial malleolus.

Bleeding time: 4 minutes. No increased capillary fragility. Histamine - fast achlorhydria with normal pepsin; Wassermann reaction etc., negative; No bilirubinaemia; Liver function tests normal; No spectroscopic evidence of urobilinuria.

BLOOD:- 1.49 million red blood cells per cu. mm.; Haemoglobin 4.6 G. per 100 ml.; Haematocrit 14%; M.C.V. 94 cu. u; M.C.H.C. 33%; M.C.H. 30.9 ; M.C.D. 8.494 u; 3,400 White cells per cu. mm.; Neutr. 59%; Lymph. 35%; Mon. 3%; Eos. 2%; Bas. 1%; anisocytosis; anisochromia; poikilocytosis with normoblasts seen in peripheral smear; reticulocytes 2%; platelets 286,000 per cu. mm.; bone marrow - megaloblastic hyperplasia; X - Ray bones : normal.

PROGRESS AND TREATMENT:- 1000 mgm. ascorbic acid given intravenously on admission then daily for four days. Mixed hospital diet given on second day after admission (fasted for fractional test meal). No other medication.

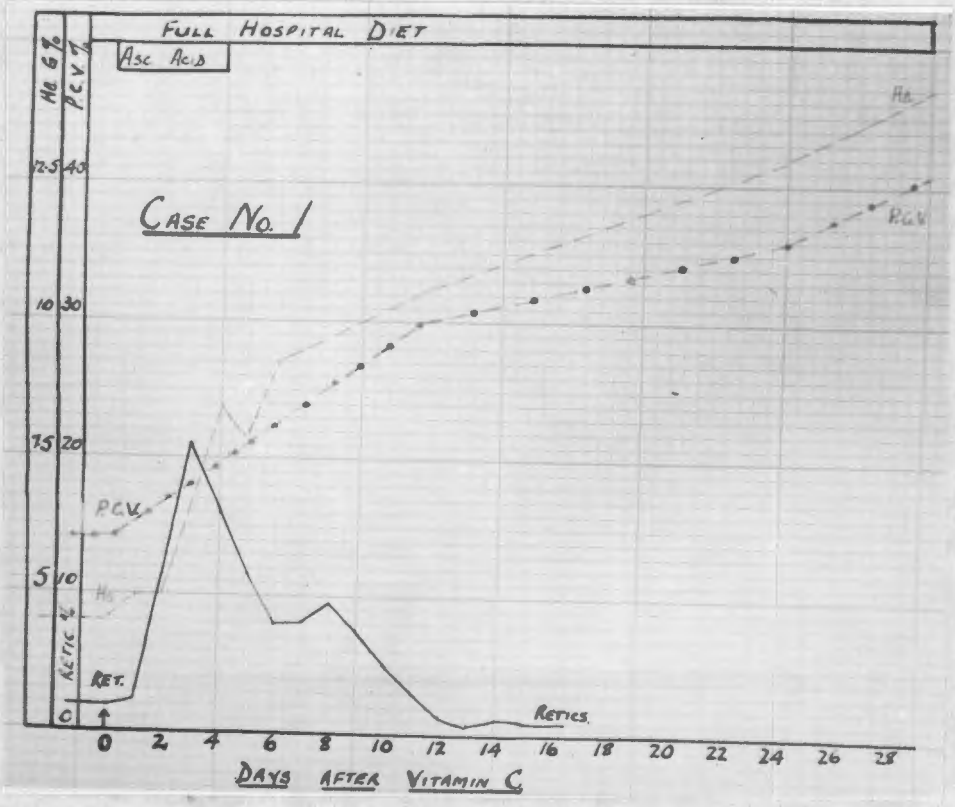
(See Graph)

Reticulocyte Peak (21%) on fourth day followed by rise of haemoglobin and red blood cells. On discharge packed cell volume was 41% (seven weeks later).

Repeated tests:- Bone marrow: normal; His-

tamine fast achlorhydria still present; Liver function tests normal (all performed after the sixth week).

Ulcer completely healed after eighteen days, gums ten days; no thickening of subcutaneous tissues or muscles, no follicular hyperkeratosis after six weeks.



PREVIOUS ADMISSION:- 1947 with weakness, joint stiffness due to "induration" in neighbouring tissues and pain with tenderness in popliteal fossae, calves, thighs and arms; Chancroid with urethral discharge.

Marked pallor 1.79 million R.B.C's per cu. mm. Haemoglobin 4.5 G %, W.B.C. 4,250 per cu. mm. Follicular

hyperkeratosis, pyorrhoea. Ten days later 2.96 million R.B.C's per cu. mm. with 8.25 haemoglobin on mixed diet and multiple vitamin therapy, iron and liver all given together. Discharged fit and well.

C A S E : 2.

No. 180075. Bantu male (Nyasa) aged 37 years; admitted on 17th May, 1949; discharged 1st. July, 1949.

HISTORY:- For two weeks he has had aching pains in the muscles of both legs and tender gums.

DIET:- Factory worker, provides and cooks his own food. Bread, mealie meal porridge, fish, coffee, for nine months. Sending money home to his family.

EXAMINATION:- Follicular hyperkeratosis, "spongy" gums, septic teeth, halitosis and pyorrhoea. Both calves slightly thickened in subcutaneous area near Tendo Achilles; no obvious large haematomata; B.P. 130/90 mms. Hg.; Splenomegaly three cms. below left costal margin, very firm in consistency; no malarial parasites detected; no pyrexia.

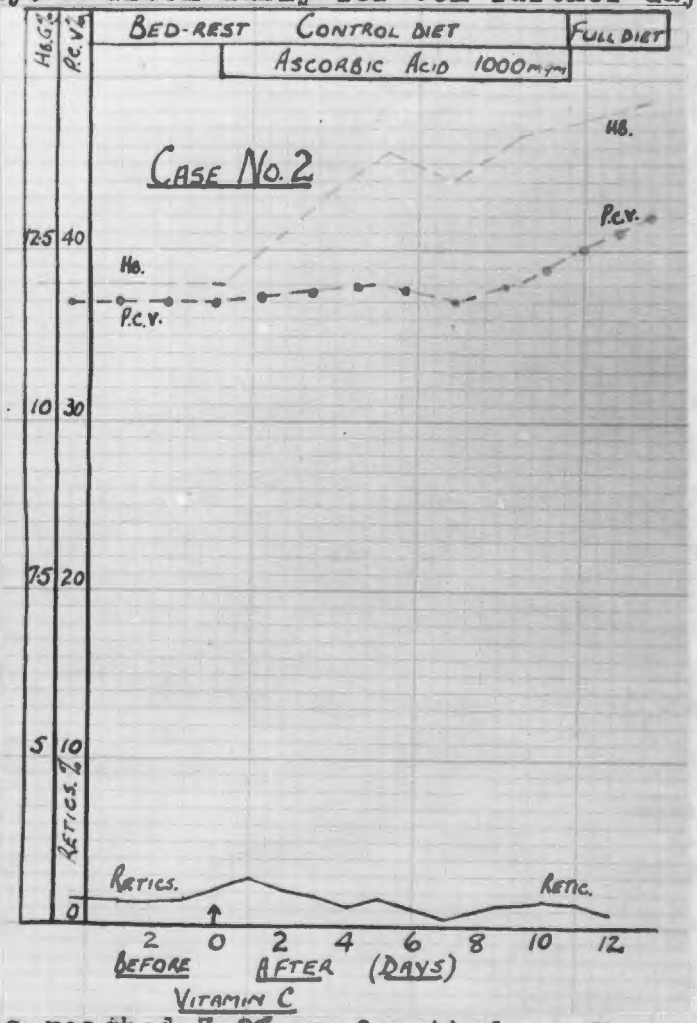
Bleeding time, capillary fragility, coagulation time, all normal.

Histamine fast achlorhydria with normal pepsin. Wassermann reaction etc., negative.

Serum van den Bergh: feeble delayed positive, serum bilirubin 3.2 mgs. %; Serum Proteins 8.0 G % with serum albumen 4.1 G % and serum globulin 3.9 G %; serum cholesterol 81 mgs. %, Thymol Turbidity 4 units; Colloidal Gold (serum) 4; Thymol Flocculation 3; Bile and urobilin present in the urine.

BLOOD:- 3.9 million red cells per cu. mm.; Haemoglobin 14.0 G.; Haematocrit 37%; M.C.V. 94.8 cu. u; M.C.H.C. 37.9 %; M.C.H. 35.9 ; M.C.D. (Diffraction method) 7.8 u; 7,040 white cells per cu. mm.; Neutr. 48%; Lymph. 50%; Mon. 2%; Smear normal; Reticulocytes 1.2 %; Sedimentation Rate 8 mm. per hour (West.); Platelets 210,000 per cu. mm.; Bone marrow: normal.

PROGRESS AND TREATMENT:- Placed on same diet as received outside. On third hospital day given 1000 mgms. ascorbic acid intravenously. Given daily for ten further days.



Reticulocytes reached 3.2% on fourth day after ascorbic acid.

Packed cell volume reached 40% after eleven days of treatment. Full diet then given with no further rise in packed cell volume.

Became "saturated" after four days of 1000 mgm. ascorbic acid.

"Spongy" gums decreased within five days of treatment, foetor being first to disappear. Some gingival hypertrophy still present three weeks later but quite marked dental sepsis present.

Splenomegaly remained unchanged. Malarial parasites never isolated, no pyrexial episodes or history of such attacks. Explained as inactive chronic malarial spleen.

Follicular hyperkeratosis was last feature to go (approximately six weeks after treatment).

Histamine-fast achlorhydria persisted with alkaline phosphatase at upper limit of normal on discharge.

C A S E : 3.

No. 180074. Bantu male (Nyasa) aged 40 years; admitted on 17th May, 1949; discharged 1st. July, 1949.

HISTORY:- Over the last two months his gums have been painful and have occasionally oozed a little blood. The lower teeth have become loose.

Pain just preceded swelling in the region of the right knee two weeks ago. This pain is much worse on moving.

DIET:- Factory labourer, lives in a compound where he has to provide and cook his own food - mealie meal porridge, bread, occasionally fish and tea without milk. Sends money home to his family.

EXAMINATION:- Pale mucous membranes, follicular hyperkeratosis "spongy" gums, dental sepsis, halitosis, loose lower incisors; moderate haematoma in upper part of popliteal fossa extending up into thigh.

Splenomegaly two and a half cms. below left costal margin, very firm in consistency.

Tachycardia, collapsing pulse B.P. 105/50 mms. Hg. Systolic praecordial murmurs. Fundus oculi : some pallor.

Bleeding time, capillary fragility, coagulation time, all normal.

Histamine - fast achlorhydria with normal pepsin.

Wassermann reaction etc., negative.

Serum Bilirubin slight trace; Serum Proteins 6.3 G. % with albumen fraction 3.5 G. and globulin 2.8 G., Thymol Turbidity 1.5 units; Colloidal Gold 1; Thymol flocculation 1; Serum Cholesterol 189 mg. %; Urobilinuria (spectroscope); trace albumenuria.

BLOOD:- 1.97 million red cells per cu. mm.; haemoglobin 6.0 G.% haematocrit 20%; M.C.V. 101.2 cu. u; M.C.H.C. 30%; M.C.H. 30.3 ; M.C.D. (diffraction method) 7.85 u.; White cells 8,600 with Neutr. 55%; Lymph 45%; Smear : Anisocytosis, anisochromia, poikilocytosis slight; Reticulocytes 1%; Sedimentation Rate uncorrected 120 mms. per hour (West.) - corrected 24 mms. per hour; Platelets 302,000 per cu. mm.; Bone marrow : normoblastic hyperplasia.

PROGRESS AND TREATMENT:- Kept on same diet (mealie meal porridge, bread and tea). On third day given 1000 mgm. ascorbic acid intravenously and then daily for ten days. Maximum reticulocyte response of 13% on sixth day followed by rapid rise in packed cell volume and haemoglobin.

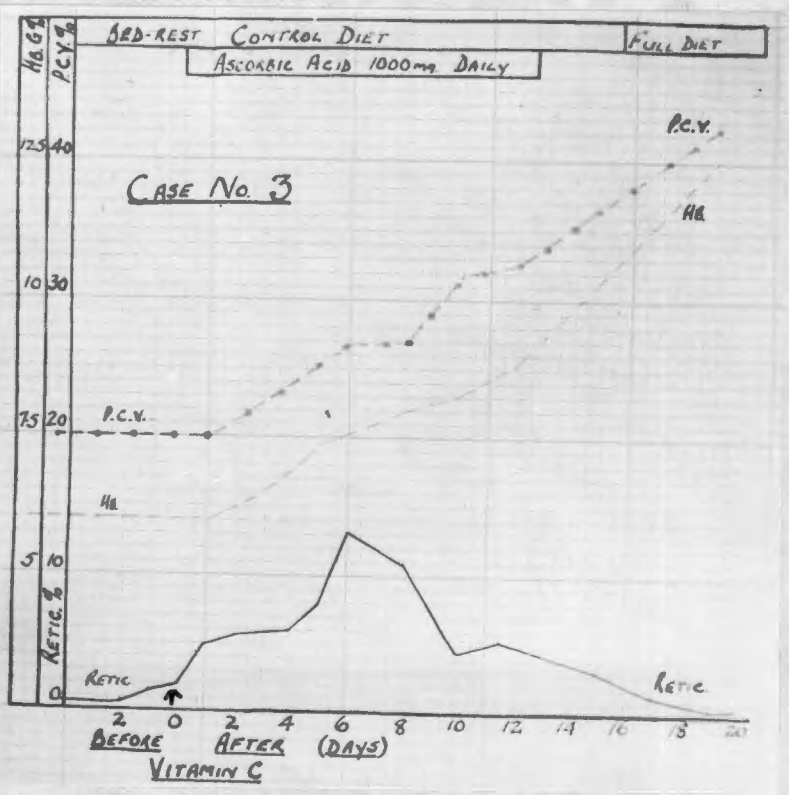
(See Graph)

Full diet given with no change haematologically (42% packed cell volume).

Became "saturated" after five days of 1000 mgm. daily.

Dental sepsis persisted but "spongy" gums had disappeared by twelfth day of treatment. Follicular hyperkeratosis no longer present at end of fifth week.

Residual non-tender thickness of popliteal fossa still present in fourth week. Completely gone by seventh week.



Splenomegaly did not change. Considered to be (like Case 2) a chronic inactive malarial spleen.

On discharge : serum protetins 7.0 G. % with albumen fraction 4.9 G. and globulin 2.1 G.%; Thymol turbidity 3 units; Colloidal Gold 2; Thymol flocculation 1; Blood urea 26 mg.%.

C A S E : 4.

No. 187465. Bantu male (Xosa) aged 49 years; admitted 30th August, 1949; discharged 4th October, 1949.

HISTORY:- For the last week he has been troubled by a pain which has spread up from his ankle to the calf and behind the right knee, much worse on walking. Swelling has developed over this time in this region.

DIET:- Cement-mixer, provides and cooks his own food - mealie meal porridge, brown bread, "samp" mealies, meat every other day, tea without milk. Very occasionally buys vegetables, cuts them up well and boils them with the meat as a stew. Boils them until the meat is cooked. Seven months duration of this diet.

EXAMINATION:- Slight interdental hypertrophy, "ooze" blood on pressure, follicular hyperkeratosis on anterior aspects of both thighs. Very large haematoma right calf and popliteal fossa, pitting oedema of skin over anterior aspect of right leg and small serous (on aspiration) effusion of right knee. B.P. 135/85 mms. Hg.

Serum bilirubin negative; serum proteins 6.0 G.% with albumen 3.8 G and globulin 2.2 G.; Thymol turbidity 1.5 units, Colloidal Gold 1, Thymol flocculation 0.

Achlorhydria with free acid of 17 units after histamine; pepsin normal.

Serum calcium 9.3 mg.%.

BLOOD:- Red cell count 2.2 million per cu. mm.; Haemoglobin 7.0 G.%; Haematocrit 23%; M.C.V. 106 cu u; M.C.H.C. 30.2%; M.C.H. 31.8 ; M.C.D. 7.979 u; White cell count 6,500 with Neutr. 65%; Lymph 20%; Mon. 5%; Reticulocytes 8%; Smear : anisocytosis of moderate degree; Platelets 235,000 per cu. mm.

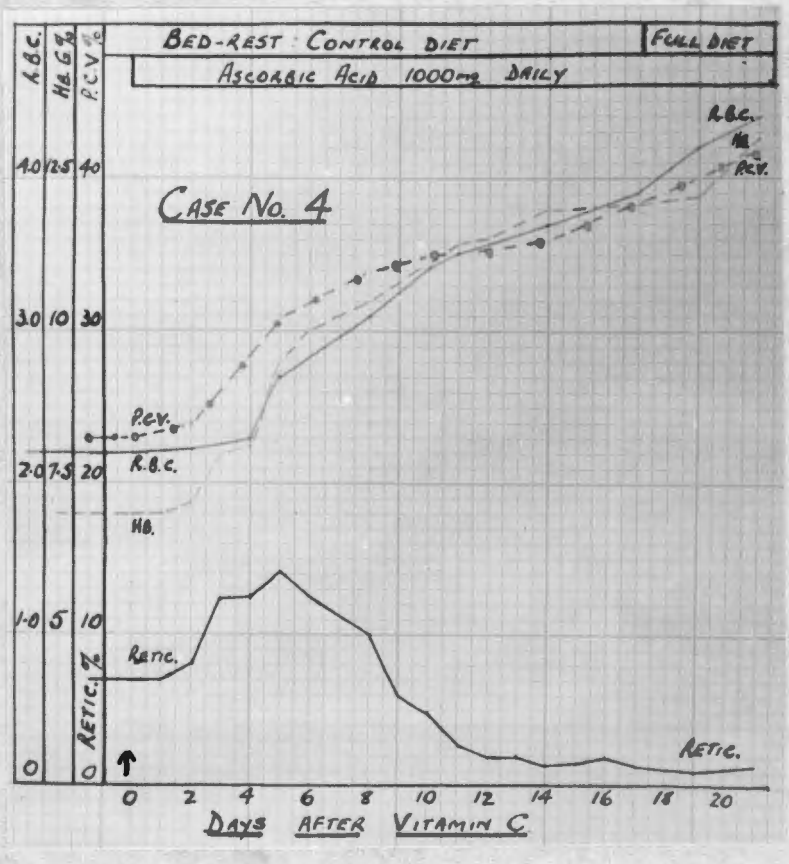
Bone marrow:- Normoblastic hyperplasia, few megaloblasts (0.8%) seen.

PROGRESS AND TREATMENT:- Placed on same diet as received outside except that meat and vegetables withheld. Given 1000 mg. ascorbic acid intravenously from the day of admission to the twentieth day. Became "saturated" in five days. Reticulocytes rose to 14% on fifth day. Put on full diet on sixteenth day after treatment when packed cell volume was 38%. Packed cell volume 42% on discharge.

(See Graph)

By twentieth day all clinical signs had disappeared except follicular hyperkeratosis which was just detectable.

Given vitamin B complex by injection on twenty-seventh hospital day with no further rise in packed cell volume or haemoglobin.


BED-REST CONTROL DIET
FULL DIET
ASCORBIC ACID 1000mg DAILY
CASE No. 4
R.B.C.
HB.
P.C.V.
RETIC.
R.B.C.
HB.
P.C.V.
RETIC.
RETIC.
4.0
3.0
2.0
1.0
0
12.5
10
7.5
5
0
40
30
20
10
0
0 2 4 6 8 10 12 14 16 18 20
DAYS AFTER VITAMIN C

C A S E : 5.

No. 198756. Bantu male (Xosa) aged 25 years; admitted 1st. February, 1950; discharged 28th February, 1950.

HISTORY:- Three weeks previously he had experienced pain in his limbs but this passed off after some days rest in bed. Four days previously there was a spontaneous onset of bleeding from the left nostril which was not, he said, very severe but at that time he felt giddy on assuming the erect position. He had noticed bleeding from his gums too.

DIET:- Labourer - provides and cooks his own food; thick mealie meal porridge, "thick" milk, samp, dry bread, meat twice a week (well cooked) over the previous year.

EXAMINATION:- Skin dull and dry, follicular hyperkeratosis, "spongy" bleeding gums, dental sepsis; Tachycardia B.P. 126/68; Praecordial systolic murmurs; no haematomata; slight pyrexia.

Histamine - fast achlorhydria; Wassermann reaction, etc., negative.

No bilirubinaemia; serum proteins 5.9 G.% with albumen 3.3G and globulin 2.6 G.; thymol turbidity 5.5 units; Colloidal Gold 5; thymol flocculation 4; Urobilinuria.

BLOOD:- Red blood cells 2.75 million per cu. mm.; Haemoglobin 8.2 G.%; Haematocrit 22.3%; M.C.V. 81.2 cu. u; M.C.H.C. 36.8%; M.C.H. 30 ; White blood cells 12,200 with

Neutr. 69.5%, Lymph. 22%, Mon. 6%, Eosin 2.5%; Smear : anisocytosis of marked degree, anisochromia with few normoblasts seen in peripheral smear; Reticulocytes 1.3%; Sedimentation rate 40 mm. (West) per hour uncorrected ; Platelet count 350,000 per cu. mm.; bleeding time, coagulation time and capillary fragility normal.

Bone marrow : Normoblastic hyperplasia.

PROGRESS AND TREATMENT:- Put on diet of mealie meal porridge, "samp" mealies, bread and tea without milk, no meat given and kept in bed. Nostril was plugged to prevent further haemorrhage. Reticulocytes increased up to 6.5% by fifth day, began to fall again by tenth day. After an initial deterioration in blood picture as a whole, slight regeneration occurred (packed cell volume rose 3% in all over ten days). On tenth day 1000 mgm. ascorbic acid added intravenously, and given daily for eighteen days. In the next ten days packed cell volume had risen 14% with a reticulocytosis of 14.3% on fourth day of treatment . During this period the patient was not confined to bed to see if this would retard progress.

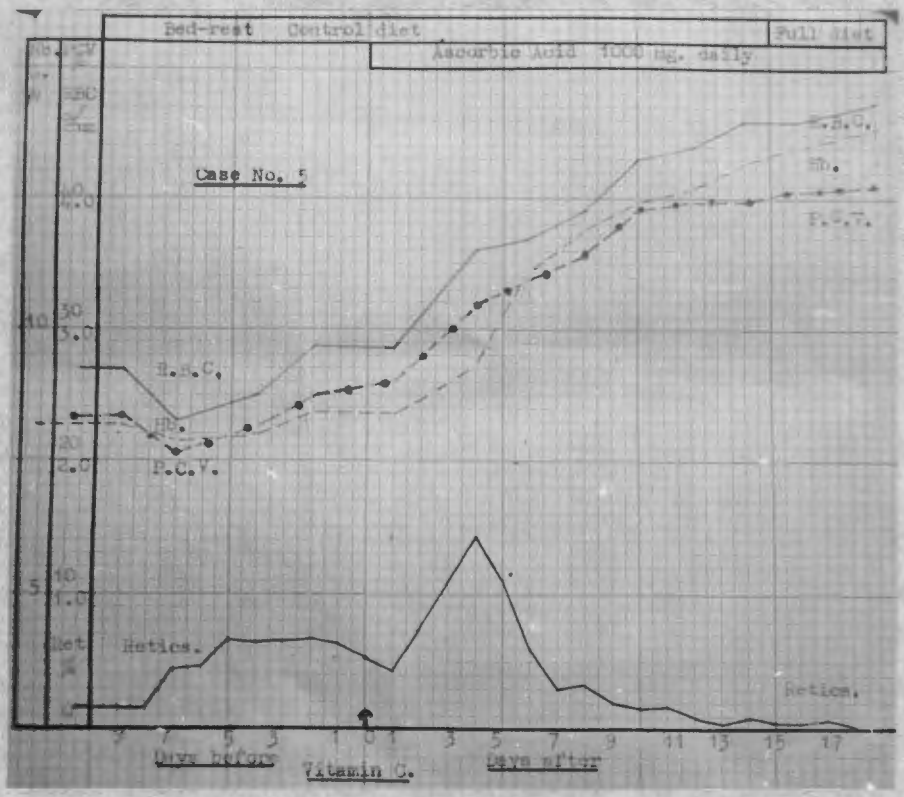
(See Graph)

Full diet with vitamin B complex given when packed cell volume was 39%. Eight days later packed cell volume had risen only 1%.

Became "saturated" after six days of 1000 mgm. ascorbic acid.

Gums had returned to normal in one week. Plug was removed from nostril forty-eight hours after admission before vitamin C was given with no further bleeding.

Achlorhydria again but free acid reached 14 units after histamine just prior to discharge.



Serum proteins 7.0 G. with albumen 4.5 G. and globulin 2.5 G.%; thymol turbidity 4.5 units; Colloidal Gold 5; thymol flocculation 4 just prior to discharge.

Bone marrow : normal.

C A S E : 6.

No. 201946. Bantu male (Nyasa) aged 27 years; admitted 11th March, 1950; discharged 19th May, 1950.

HISTORY:- Whilst carrying a very heavy weight seven weeks previously he bumped his left leg and arm. A swelling appeared on his left forearm. One week later the left calf and knee became swollen with pain on movement. He was put off work and went to bed. After two weeks in bed these swellings subsided somewhat but the leg was painful on movement or pressure.

Has never bled from his gums but has noticed pain and ulceration for two weeks. He cannot now stand erect as this causes unconsciousness.

Entered the Union of South Africa in January, 1949.

DIET:- Mixes cement, provides and cooks his own food but over the last six weeks he had to rely on the kindness of his friends as he was in bed. This meant just mealie meal porridge, occasionally bread and tea without milk and not the meat twice a week, daily bread with occasional jam - he thinks plum jam - that he had consumed as well over the last year.

EXAMINATION:- Dull, dry skin (paler colour than usual East African) gross pallor, pyrexial, perifollicular haemorrhages with pigmentation especially noticeable on dorsum wrists, pigmented area overlying some thickening in underlying muscles

on volar aspect of left forearm (the site of the former swelling). Follicular hyperkeratosis anterior aspect of both thighs. Small effusion into left knee joint, some tender thickening of both calves. No large haematomata. "Splinter" haemorrhages of finger nails; Splenomegaly 1½ cms., very firm; Tachycardia, praecordial systolic murmurs, B.P. 130/70 mms. Hg., Fundus oculi pale with a small retinal haemorrhage.

"Spongy" gums with small ulcers which can be made to bleed easily; halitosis.

Swab from mouth "Staphylococcus Aureus".

Histamine - fast achlorhydria with normal pepsin activity; no bilirubinaemia; serum proteins 7.0 G.% with albumen 4.0 G. and globulin 3.0 G.; Thymol turbidity 1 unit, Colloidal Gold 0; thymol flocculation 0; Blood urea 28 mg.%; serum calcium 10.4 mg.%; Plasma ascorbic acid 0.35 mg.%. Urobilinuria, urinary urobilinogen 37 mg. per day, faecal urobilinogen 184.5 mg./100 G.

Bleeding time, coagulation time, red cell fragility, normal. Capillary fragility test led to large haematomata down forearm but no petechiae.

BLOOD:- Red blood cells 1.83 million per cu. mm.; haemoglobin 5.4 G.%; haematocrit 15%; M.C.V. 82 cu. u; M.C.H.C. 36%; M.C.H. 30 ; M.C.D. 6.97 u; White blood cells 3,900 with

Neutr. 44.5%, Lymph. 49.5%, Mon. 4%, Eosin 2%; Smear : marked anisocytosis, and anisochromia, poikilocytosis; Reticulocytes 4.1%.

Bone marrow : intense normoblastic hyperplasia with very few mitotic figures seen.

Plasma Iron and Iron Binding capacity low [Budtz Olsen (1950)(a)].

PROGRESS AND TREATMENT:- Confined to bed on mealie meal porridge, dry bread, "samp" mealies, "black" tea. During this time several tests were carried out. Various haematinics were given and the effect on the blood and bone marrow observed. Liver function tests and serum protein estimations were repeatedly made. There was no significant change. The faecal and urinary urobilinogen in accurate quantitative studies showed that by causing fresh haematomata, a rise in both faecal and urinary urobilinogen occurred without bilirubinaemia or change in the red, white cells or reticulocytes.

After a week's bed rest alone the following were tried at approximately weekly intervals. Vitamin B 12 30 u G. daily for four days by injection, Folic acid 15 mgm. a day for five days by mouth, Iron as "Ferrivennin" in doses of 400 to 500 mgm. intravenously over five days. Despite all this the reticulocyte level remained more or less constant at 6 or 7% and the packed cell volume slowly fell.

The mild pyrexia persisted, having been at its highest (101°F.) just after the production of the new haematomata. The urinary output dropped despite a more or less constant fluid intake.

After approximately thirty days 1000 mgm. ascorbic acid was given intravenously and thereafter daily. A reticulocytosis of 20% occurred on the fifth day of treatment with rapid haematological regeneration. The packed cell volume doubled itself in nine days, was more than 30% in a fortnight and reached 40% from an initial level of 12.5% in four weeks.

(See Graph)

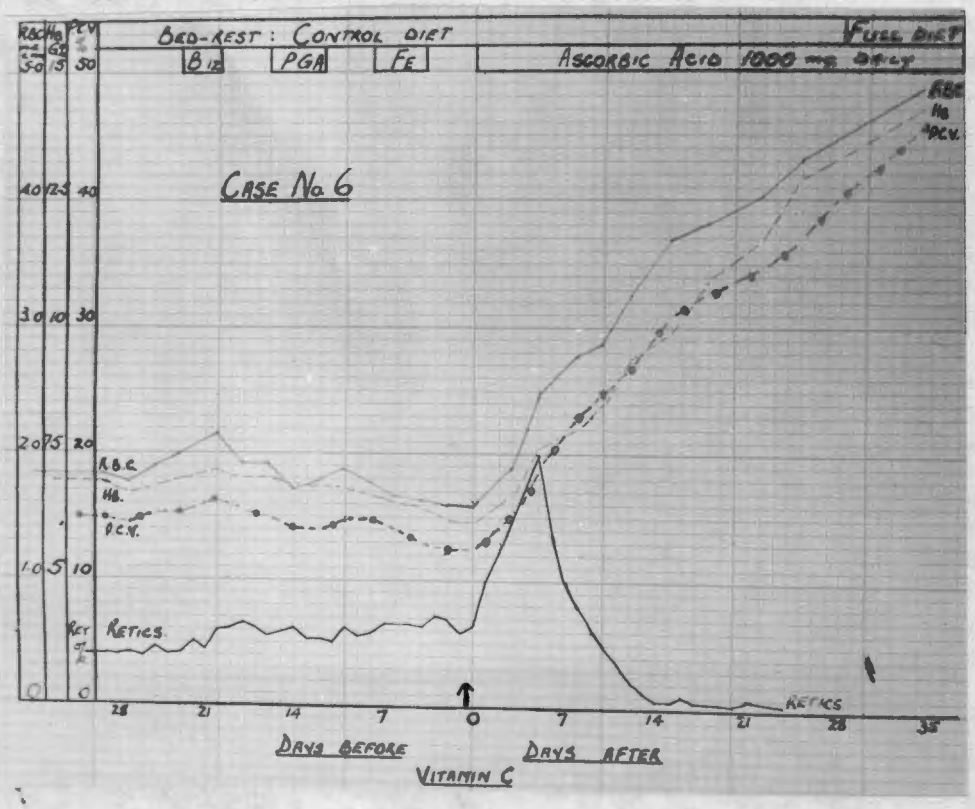
Only then was a full hospital diet given.

The bone marrow estimations done weekly to determine whether any haematinic given would cause a change, showed the same picture of intense cellularity mainly at the basophilic normoblast stage with the striking infrequency of mitotic figures prior to ascorbic acid treatment. Two weeks later the picture was one approaching normal with numerous mitotic figures.

There was no change in the haematological indices (M.C.V., M.C.H.C.) or the M.C.D. as measured by the diffraction method. The picture remained normocytic and normochromic throughout.

The plasma iron reached the lower limit of normal after four weeks treatment.

The sedimentation rate after its initial rise rapidly dropped to normal as the anaemia improved. The corrected sedimentation rate initially was 24 mm. per hour (West.) The pyrexia likewise dropped to normal.



The white blood cells after remaining between 3,900 and 4,200 per cu. mm. before treatment rose to and remained between 6,000 and 7,000 per cu. mm.

The urinary urobilinogen dropped rapidly as the anaemia improved but the faecal level dropped much slower as the haematomata were slowly absorbed.

The gums: coagulose positive staphylococcus aureus was isolated on admission. During the first week penicillin lozenges were given. Pain and halitosis improved but the gingival hyperplasia only returned to normal after a week of vitamin C.

Just prior to treatment the serum globulin had risen 0.2 G. to 3.2 G.% out of a total of 7.0 G.%. Two months after treatment it was 2.7 G.% out of a total of 7.3 G.%. Liver function tests showed no significant change.

Five weeks after treatment, no anaemia being now present, the achlorhydria was still histamine - fast. A barium meal examination showed a deficiency small bowel pattern.

The urinary output improved steadily after ascorbic acid therapy.

The skin became shiny, first commented upon by the patient, the pigmented areas gradually faded. No residual muscle thickening was palpable at six weeks. The follicular hyperkeratosis disappeared at the same time.

C A S E : 7.

No. 58246. Bantu male (Zulu) aged 50 years; admitted 30th March, 1950; discharged 19th May, 1950.

HISTORY:- For the previous three weeks the patient became progressively more weak so that he had difficulty in walking or standing. Due to a painful mouth he had difficulty in talking or eating. He has lost weight.

DIET:- Out of work for three months. Often goes without food for two days. Mealie meal porridge and "black" tea occasionally over the last month. No bread for two months.

EXAMINATION:- Drowsy, unco-operative, so weak that he cannot sit up, confused with continual involuntary movement of his lips, arms and hands. "Sharkskin" nasal sebaceous plugging, conjunctival injection and vascularisation of cornea (other eye was enucleated following trauma five years before), smooth magenta coloured tongue, dry, thick, rough scaly skin anterior aspect of legs and ankles; Perifollicular petechiae with follicular hyperkeratosis; "spongy" gums with halitosis.

B.P. 180/140 with some dilatation of ascending aorta on X-Ray. Wassermann reaction negative but Berger doubtful. Free acid present before (6 units) and after histamine (40 units) in the fractional test meal.

Serum Bilirubin 0.5 mg.%; serum proteins 8.5 G. with albumen fraction 5.3 G.% and globulin 3.2 G.%; Thymol

Turbidity 4.5 units; Colloidal Gold 4; Thymol flocculation 4; Blood urea 40 mg.%; Urinary urobilinogen 19.5 mg./day; faecal urobilinogen 120 mg./day.

BLOOD:- Red cells 6.35 million per cu. mm.; Haemoglobin 18 G; Packed cell volume 59%; White cells 7,150 per cu. mm. with 51% Neutrophils, 37% lymphocytes, 10% monocytes and 2% Eosinophils.

PROGRESS AND TREATMENT:- Placed on mealie meal porridge, bread "black" tea and "samp" mealies. Vitamin B complex as "Bejectal" 2 ccs. daily was given intramuscularly with cure of all signs associated with this deficiency. Remarkable improvement mentally but left with "spongy" gums and peri-follicular haemorrhages.

Used thereafter as a control.

With the rest in bed the urinary urobilinogen slowly dropped but remained above normal throughout or at the upper limit of normal. Faecal urobilinogen remained at low normal. An attempt to produce haematomata with a blood pressure cuff failed. Forty ml. of blood was removed from an arm vein and injected into the buttock. The urinary urobilinogen rose from 4 mg. per day to 12 mg. per day, while the faecal urobilinogen rose from 20 mg. per 100 G. to 96 mg. and 110 mg. per 100 G. two and three days later with a slow fall to 42 mg. per 100 G. after another four days.

Forty ml. of blood was then removed and discarded

to see whether the removal of this amount would cause effects. No change occurred.

The reticulocytes remained unchanged at a level of less than 1% throughout. The sedimentation rate remained between 3 and 8 mm. (West.).

One month after admission the serum proteins were 7.3 G. with albumen 4.6 G. and globulin 2.7 G.%; Thymol Turbidity 7 units; Colloidal Gold 3 and Thymol flocculation 3.

A repeat blood count showed 6.03 million with haemoglobin 17 G., packed cell volume 53% with white cells 6,500 per cu. mm. This was considered insignificant as the question of dehydration causing the high initial figures had not originally been considered.

Vitamin C intravenously in 1000 mgm. doses was given after five weeks.

The gums became normal first with slow fading of the perifollicular haemorrhages.

There was no alteration in urinary output which was normal before.

The fractional test meal was unchanged essentially, with free acid before and after histamine.

C A S E : 8.

No. 205796. Bantu male (Xosa) aged 35 years; admitted 21st April, 1950; discharged 14th July, 1950.

HISTORY:- For the five weeks previous to admission he has noticed a painful mouth with swollen gums which did not bleed. His tongue was not painful.

For approximately the same duration the left calf has been very swollen and tender to the touch. The knee and ankle movements have been hindered by this. For the same time there has been a similar swelling of the right forearm; no associated trauma.

He does not think he has lost weight but he has had for a few months a slight cough.

DIET:- Quarry worker, provides and cooks his own food; mealie meal porridge, black tea, occasionally sour milk; no meat-for six years.

EXAMINATION:- Pallor, follicular hyperkeratosis of anterior aspect of both thighs. Haematoma of right forearm; haematoma of left calf, warm and tender with overlying glossy skin. Dental caries, swollen blueish-red gums with swelling extending on to palate; halitosis. B.P. 105/60 mms. Hg. with praecordial systolic murmurs. Pyrexia.

Histamine - fast achlorhydria with normal pepsin activity. Serum van den Bergh direct negative with a very

slight trace of serum bilirubin with icteric index 14; serum proteins 5.9 G. with albumen 3.4 G. and globulin 2.5 G. per 100 ml. Thymol Turbidity 2.5; Colloidal Gold 1; Thymol Flocculation 1; Urine Urobilinogen 59 Ehrlich Units per day; Faecal urobilinogen 340 mg. per 100 G.; serum cholesterol 92 mg.%.

Serum Calcium 7.8 mg.% (serum albumen 4.1 mg.% at that time); Blood urea 36 mg.%. Wassermann reaction etc., negative.

BLOOD:- 2.69 million red cells per cu. mm.; 7.3 G. per 100 ml. haemoglobin; haematocrit 22.5%; M.C.V. 84 cu. u; M.C.H.C. 32.4%; M.C.H. 27γγ; M.C.D.7.02 u. White blood cells 7,200 per cu. mm. with Neutr. 65%; Lymph. 32%; Monocytes 3%. Peripheral smear: marked anisocytosis, anisochromia. Reticulocytes 7.4%; platelets 243,000 per cu. mm.; sedimentation rate 60 mm. per hour (West.).

Bleeding time, coagulation time normal.

Bone marrow: normoblastic hyperplasia with preponderance of basophilic normoblasts and mitosis infrequently seen.

X - Ray chest and bones of legs and forearms showed thickened apical pleura in the chest and normal bones.

PROGRESS AND TREATMENT:-

Placed in bed on the same diet of mealie meal porridge and black tea. Urinary urobilinogen slowly fell to reach the upper limit of normal by the fourteenth day. The faecal urobilinogen slowly climbed and was 612 mg. per 100 G. at that time, and then slowly dropped.

The reticulocytes remained at about the same level and despite this the blood count fell to 19.8% Haematocrit where it remained until the tenth day. Then it slowly returned to its initial level until just before treatment with ascorbic acid. Folic acid, vitamin B12 and vitamin B complex had no effect. The haematological indices remained unchanged.

The plasma iron dropped from its initial level of 56 u G. per 100 ml. to 44 u G. and on the day after haematomata were produced with a blood pressure cuff it was 18 u G. per 100 ml. The production of these haematomata caused a very marked rise in faecal urobilinogen from 210 mg./100 G. to 1139.7 mg./100 G. with normal urinary urobilinogen levels as performed with petroleum ether extraction. The icteric index at this time was 22.4 units with a serum bilirubin of 0.6 mg.%.

Until this time coincident with the drop in urinary urobilinogen the serum globulin fell from 2.5 G.% to 1.5 G.% whilst the albumen fraction rose from 3.4G. to 4.1 G.% The Thymol flocculation became 0, but the Colloidal Gold rose from 1 to 3 units whereas the Thymol Turbidity remained at 2.5 units.

The bone marrow differential count showed the same normoblastic hyperplasia with an increase in the percentage of basophilic normoblasts which were present in "clumps" or "nests" throughout.

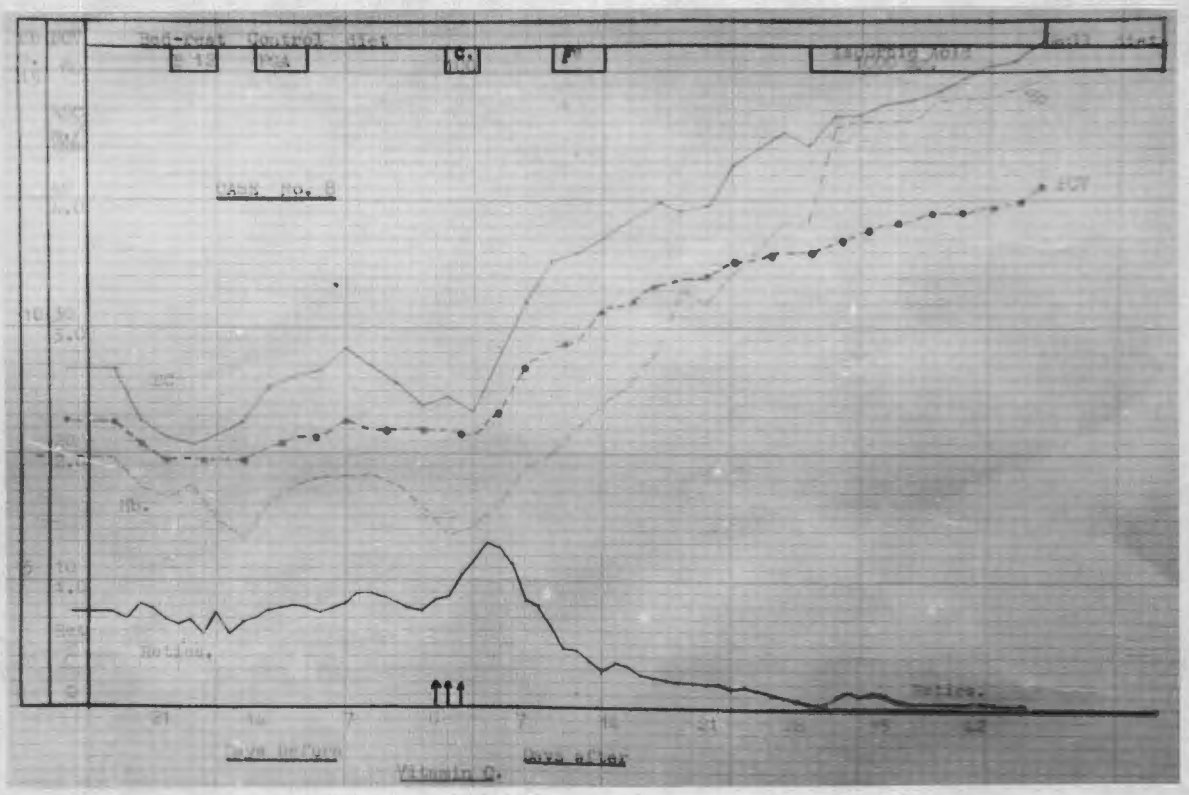
In this case a change in procedure was adopted. It was decided to give only 300 mg. ascorbic acid divided over three days by mouth, observe if a response occurred and then to give iron intravenously to see if a second response occurred. In this way it may have been shown that the failure of response to iron in other cases was due to the absence of vitamin C - a factor which may have been necessary for its utilisation. From the accompanying graph it will be seen that this small dose of ascorbic acid by mouth caused a rise in reticulocytes to 13% on the fifth day followed by a rise in packed cell volume of 1% per day. Iron as "Ferrivennin" was given as the reticulocytes were falling with no subsequent rise or "double response". Furthermore, the rate of regeneration slowed down. The packed cell volume had increased 10% in the first ten days after ascorbic acid therapy but in the following ten days it moved up only 2%.

For the subsequent ten days (i.e. 20 to 30 days after ascorbic acid) it remained stationary at 36%. Ascorbic acid (1000 mg.) was then given intravenously. In a fortnight the blood picture was normal and full diet was given. Bone marrow examination at that time was normal.

The pyrexia, which had become more marked at the

time when more haematomata were produced, disappeared after the first dose of ascorbic acid. Coincident with this was an increased urinary output.

The histamine - fast achlorhydria was still present six weeks after therapy had been given.



The clinical features responded to oral ascorbic acid, the gums rapidly, but the thickening from the old haematoma in the calf was still just palpable on discharge two months later. The halitosis and obvious dental sepsis, but not the gingival hyperplasia had already cleared on antiseptic mouth washes during the first week of admission.

The faecal urobilinogen slowly dropped to normal. The serum proteins rose from 5.5 G. per 100 ml. to 6.9 G. per 100 ml. with the albumen fraction being responsible i.e. rising from 3.4 G. to 4.8 G. per 100 ml. The urinary urobilinogen remained normal but the other liver function tests remained disturbed.

The plasma iron had risen to 56 u G. per 100 ml. from the initial 18 u G. prior to ascorbic acid. This reading was taken five days after the first dose of ascorbic acid. Two weeks later it was 86 u G. per 100 ml. and reached 393 u G. per 100 ml. five minutes after iron intravenously. At this time the packed cell volume was stationary at 36%.

There was a slight increase in weight when ascorbic acid alone was added to the diet but a marked increase occurred once full diet was given.

C A S E : 9.

No. 207166. Bantu male (Nyasa) aged 24 years; admitted 9th May, 1950; discharged 14th July, 1950.

HISTORY:- For four weeks he has had swollen and "spongy" gums which did not bleed. At the same time his mouth has been so painful that despite a good appetite he is afraid to eat because of pain. Two weeks ago he developed spontaneously a diffuse swelling of the right calf. This has been very painful, especially on walking. It has felt warm and causes difficulty in bending the knee. He has been confined to bed for the last ten days. Just prior to the onset of his illness he had an episode of diarrhoea.

DIET:- Cement-worker, provides and cooks his own food. Mealie meal porridge, brown bread, dried small brown beans, black tea, occasional sour milk, dried fish and rarely meat. Duration of diet: eleven months.

EXAMINATION:- Very dark skinned with gross pallor of mucous membranes except the gums which are swollen overlapping the teeth in places and blueish red in colour. Halitosis. Follicular hyperkeratosis of thighs sparing the inner aspects. Very slight sebaceous nasal plugging ("Sharkskin"). Large haematoma (intramuscular) of calf and thigh of right lower limb. Skin over it glossy, warm with slight pitting oedema anteriorly over the tibia. Fluid in right knee joint. B.P.130/75 mms. Hg. Pyrexial.

Histamine-fast achlorhydria with normal pepsin activity; Serum van den Bergh and bilirubin negative; serum proteins 7.0 G.% with albumen fraction 4.6 G. and globulin 2.4 G. %. Thymol turbidity 4 units; Colloidal Gold 2; Thymol flocculation 1; Serum Cholesterol 109 mg.%; Urinary urobilinogen 16.2 Ehrlich units per day; Faecal urobilinogen 284 mg./100 G.; Serum calcium 10 mg.%; Wassermann reaction negative; Plasma ascorbic acid 0.3 mg. %.

BLOOD:- Red blood cells 2.8 million per cu. mm.; Haemoglobin 7.3 G.%; Haematocrit 22.8%; M.C.V. 81.4 cu. u; M.C.H.C. 32%; M.C.H. 26% M.C.D. 7.504 u; White blood cells 8,150 per cu. mm. with Neutr. 57%, Lymph. 38%, Mon. 4%, Eosin 1%. Peripheral smear - moderate anisocytosis and "anisochromia"; Reticulocytes 1.8%; Platelets 215,000 per cu. mm. Sedimentation rate 130 mm. per hour (West.)(uncorrected).

Bleeding time and coagulation time normal.

Bone marrow: Normoblastic hyperplasia with an increase in early normoblasts.

Plasma Iron 44 u G. per 100 ml.

PROGRESS AND TREATMENT:- Placed in bed on same diet, mealie meal porridge, bread and tea. Urinary urobilinogen reached normal on twelfth day as did the faecal urobilinogen. The packed cell volume rose slightly to reach 24% on the tenth day but over the next week dropped so rapidly (5%) that it was deemed necessary to administer ascorbic acid. In the meantime

there had been no response to vitamin B12, Folic acid, vitamin B complex and "Ferrivennin". During this period the reticulocytes had risen to remain between 3% and 5%. Blood was noticed in the stools for a few days but no organisms or entamoebae were isolated. Streptomycin and sulphasuccidene had no effect on this or the pyrexia. The pyrexia disappeared once ascorbic acid was given as did the blood in the stools. At no time was this severe. The haematological indices and bone marrow remained unchanged over this period. The plasma iron, however, dropped from the initial 44 u G. to 12 u G. per 100 ml.

Intravenous ascorbic acid (1000 mg.) was then given daily (after seventeen days control period). There was a reticulocyte peak of 13% on the sixth day with the packed cell volume reaching 38% in just over three weeks having added only ascorbic acid to the basal diet. Full diet was given when the packed cell volume was 41%.

(See Graph)

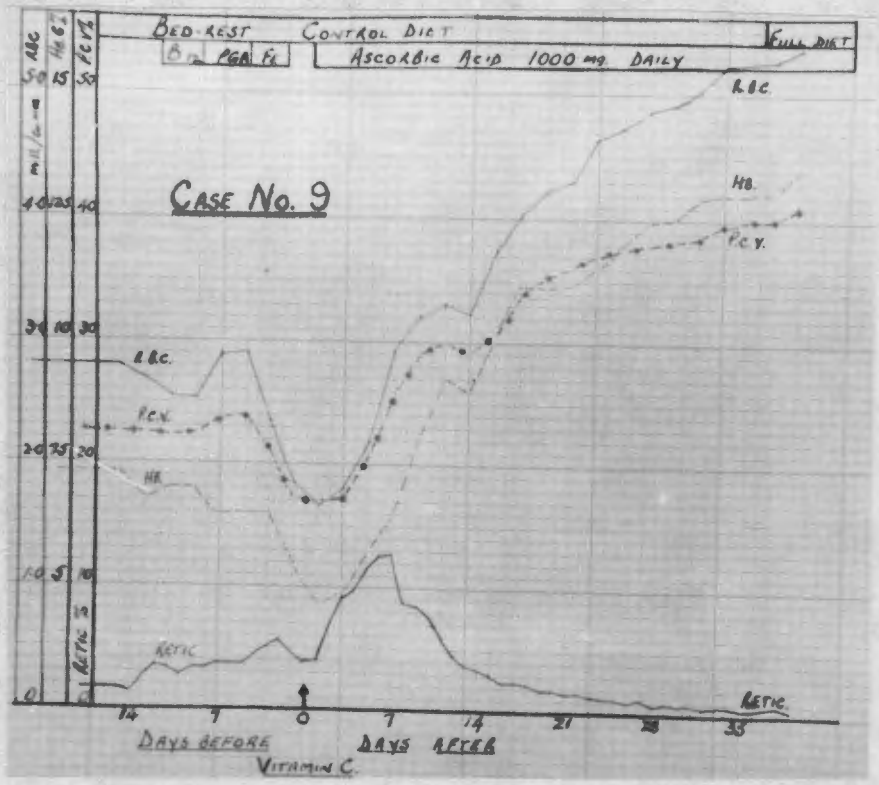
The bone marrow reverted to normal.

The clinical features rapidly responded. Considerable absorption of the haematoma had taken place, however, prior to ascorbic acid therapy.

There was an increased urinary output.

The total serum proteins increased slightly to 7.5 G. % but the albumen globulin ratio remained unchanged.

Free acid (6 units) returned and reached 13 units after



histamine. Liver function tests still showed disturbed function. The plasma ascorbic acid was recorded as 0.8 mg.%.

There was a gain in weight even before full diet was given.

C A S E : 10.

No. 207705. Bantu male (Pondo) aged 59 years; admitted 22nd. May, 1950; discharged 20th June, 1950.

HISTORY:- Seven months previously he had been admitted with similar complaints of two weeks duration and no anaemia. He received 500 mg. vitamin C intravenously daily for four days and 300 mg. three times a day by mouth. After ten days he was discharged apparently completely cured. At this time there had been complaints of pain in the legs but no palpable haematomata.

After a few months the pain in the gums returned, three weeks ago collapsed after an attack of dizziness. He noticed then swelling in left calf and later right thigh and left arm.

DIET:- He has been out of work for three months. Diet: bread, mealie meal porridge, cooked small dry brown beans and black tea since discharge seven months before.

EXAMINATION:- Pallor, follicular hyperkeratosis extensor surfaces thighs and forearms, with areas of perifollicular haemorrhages. Pyrexia. Haematomata left calf, right sub-periosteal of tibia, thigh and left forearm.

Halitosis, "spongy" gums.

Liver enlarged five cms. below ninth costal cartilage. B.P. 110/70 mms. Hg.; E.C.G. normal.

Free acid before (7 units) and after (20 units) histamine.

Serum proteins 7.3 G. with 4.5 G. albumen and 2.8 G. globulin; Thymol turbidity 2.5; Colloidal Gold 1; Thymol flocculation 0; No bilirubinaemia; urinary urobilinogen 8.06 mg./day; faecal urobilinogen 303 mg./day.

Serum calcium 9.9 mg.%; blood urea 30 mg.%; Wassermann reaction negative; X-Ray chest: right apical pleural thickening with inactive infraclavicular lesion. No acid fast bacilli were found in repeated sputum examinations and gastric juice examination.

Barium meal examination normal. Passed a large tapeworm.

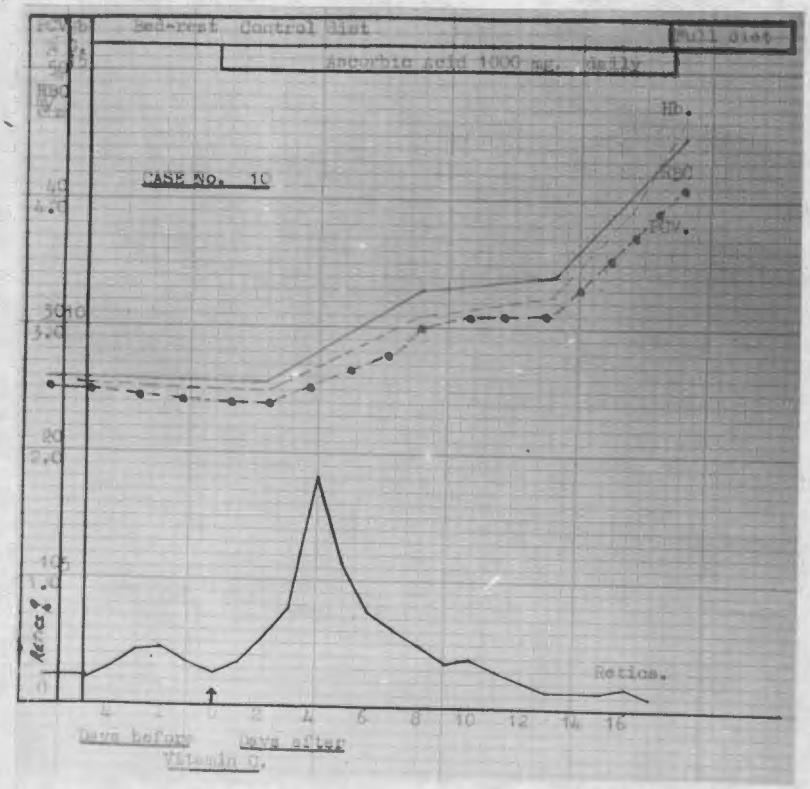
BLOOD:- Red blood cells 2.76 million per cu. mm. Haemoglobin 8.6 G.%; Packed cell volume 26%; M.C.V. 94.2 cu. u; M.C.H.C. 33%; M.C.H. 31%; M.C.D. 7.58 u; white blood cells 7,600 per cu. mm. with 46% Neutrophils, 44% Lymphocytes, 10% Monocytes. Smear: Moderate anisocytosis and anisochromia. Reticulocytes 3.3%; Platelets 270,000 per cu. mm.

Bone marrow: Normoblastic hyperplasia.

The plasma iron and total iron binding capacity were low.

PROGRESS AND TREATMENT:- Placed on mealie meal, black tea and bread diet in bed. Urinary urobilinogen fell to 2.14 mg./day. Reticulocytes reached 4.7% on the fourth day and then fell

slowly to 2.3% on the sixth day. Packed cell volume had fallen to 24%. 1000 mgm. Ascorbic acid intravenously was then given daily for ten days followed by 500 mgm. intramuscularly daily for another ten days. The reticulocytes reached a peak of 18.2% on the fourth day. Eighteen days later with packed cell volume at 41%, full diet was given.



Bone marrow was now normal.

Pyrexia disappeared on fifth day after treatment followed by improvement in the gums and other physical signs.

Thymol turbidity 5.5 units; Colloidal Gold 4; Thymol flocculation 2; Free acid (62 units) before and (78 units) after histamine on fractional test meal.

As with Cases 6, 8 and 9 increased urinary output occurred after ascorbic acid therapy.

Comment: This case shows that the incubation period of Scurvy and its anaemia is in the region of six months.

C A S E : 11.

No. 212955. Bantu male (Nyasa) aged 27 years; admitted 2nd. August, 1950; discharged 19th August, 1950.

HISTORY:- Three weeks previously the patient noticed a swelling of the right calf, which caused pain on walking.

Cleans his teeth regularly and has noticed that the gums have bled a little lately.

DIET:- Mealie meal porridge, bread, meat, cooked dry small brown beans and coffee.

EXAMINATION:- Well-nourished, follicular hyperkeratosis. Large haematoma of right calf. Gums look quite normal but on pressure blood exudes; interdental gingival hypertrophy near a carious lower left molar, but nowhere else.

Pyrexial B.P. 110/70 mms. Hg.

Free acid (10 units) with fractional test meal.

Serum bilirubin 1 mg.%; Serum proteins 6.5 G. with albumen 2.8 G. and globulin 3.7 G.%; Thymol turbidity 1.5; Colloidal Gold 0; Thymol flocculation 0; Urine urobilinogen 6.6 mg. per day; Faecal urobilinogen 265 mg. per 100 G.

BLOOD:- Red blood cells 4.3 million; haemoglobin 13.6 G.; Haematocrit 41.5%; M.C.V. 96.5 cu. u; M.C.H.C. 32.8%; M.C.H. 31.6 $\gamma\gamma$; M.C.D. 7.32 u; White blood cells 6,700 per cu. mm. with 45% Neutrophils, 45% Lymphocytes, 2% Monocytes,

7% Eosinophils and 1% Basophils; Reticulocytes 1.4%; Platelets 320,000 per cu. mm.

Bleeding time, coagulation time and capillary fragility normal; Prothrombin time normal.

Plasma Iron 28 u G. per 100 ml.; after 50 mg. Iron intravenously 54 u G. per 100 ml.

Bone marrow: normal.

PROGRESS AND TREATMENT:- Given 1000 mg. ascorbic acid with slow regression of haematoma. Ascorbic acid given daily intravenously.

After two weeks serum proteins 8.0 G.% with albumen 3.8 G. and globulin 4.2 G.%; Thymol turbidity 3.5 units; Colloidal Gold 0; Thymol flocculation 0.

Fractional test meal showed 30 units free acid before histamine and 43 units free acid after histamine.

Plasma Iron returned to normal.

C A S E : 12.

No. 212867. Bantu male (Xosa) aged 55 years; admitted (as out patient) 4th August, 1950; discharged 22nd. August, 1950.

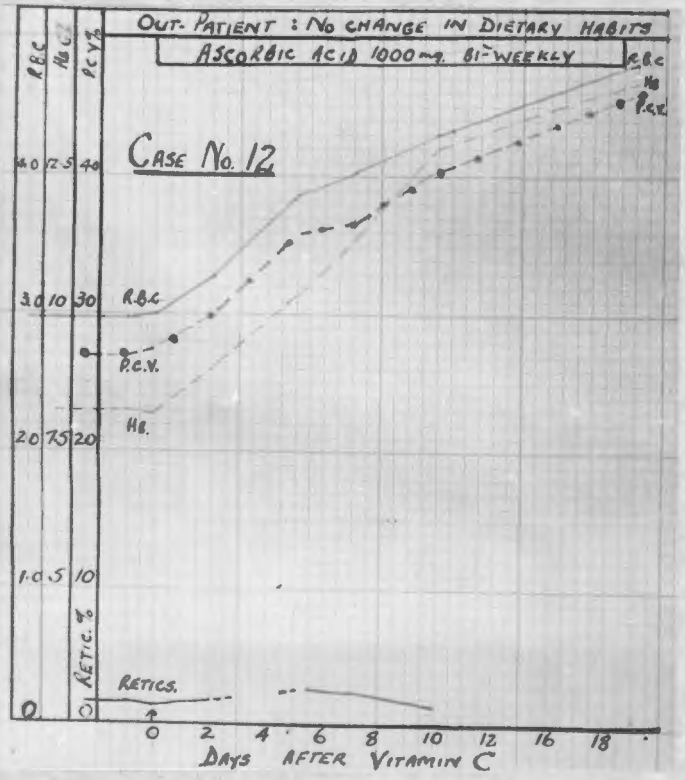
HISTORY:- Was originally diagnosed as a low grade cellulitis of the right calf as he stated that the tender diffuse swelling had occurred spontaneously one month previously and slowly increased in size.

DIET:- Dairy worker; provides and cooks his own food - mealie meal porridge, black tea and bread for nearly a year.

EXAMINATION:- Pallor with "spongy" gums and halitosis. Follicular hyperkeratosis over anterior aspects of thighs. Large haematoma of the right calf. B.P. 125/70 mm. Hg.; Serum van den Bergh direct and indirect negative; Serum proteins 7.5 G.% with albumen 3.7 G.% and globulin 3.8 G.%; Thymol turbidity 2.5 units; Colloidal Gold 1; Thymol flocculation 0; Urinary urobilinogen 1.35 Ehrlich units per 100 ml.; Faecal urobilinogen 163 mg./100 G.

BLOOD:- Red blood cells 2.9 million per cu. mm.; Haemoglobin 8.8 G.; Haematocrit 27%; M.C.V. 93.1 cu. u; M.C.H.C. 32.6%; M.C.H. 30 γγ; M.C.D. 7.185 u; White blood cells 6,000 with 61% Neutr., 30% Lymph., 8% Mon., 1% Eosin. Smear: Moderate anisocytosis; Reticulocytes 1.6%; Platelets 236,000 per cu. mm. Bleeding time, capillary fragility, coagulation time and prothrombin time normal.

PROGRESS AND TREATMENT:- There was an extreme shortage of hospital beds so it was decided to observe the patient as an out-patient. At the same time it was of interest to observe the progress with treatment while the patient was up and about. In order to check the haematological findings the patient was instructed to return the next day. The findings were essentially similar, the faecal urobilinogen was found to be 115 mg./100 G. and the Plasma Iron was low. Ascorbic acid (500 mg.) was given



intravenously and the same dose intramuscularly and the patient was instructed to consume ascorbic acid tablets of 100 mg. strength three times a day but to remain on the same diet. Each subsequent visit twice a week the blood count was performed and the same treatment repeated.

Rapid haematological regeneration followed. The packed cell volume had risen 9 % in a week and reached 46% after eighteen days of treatment. No marked reticulocytosis occurred. This was possibly due to the fact that the anaemia was not severe enough rather than the peak being missed due to the infrequent counting.

The gums rapidly improved and despite being up and about the haematoma slowly became absorbed. Residual thickening was still present one month later but the follicular hyperkeratosis had almost completely disappeared.

The plasma iron was almost normal.

At the conclusion of treatment an independent observer without realising the significance thereof was asked to take a detailed dietary history with particular reference to the last three weeks. Adherence to the original diet had been maintained. The patient was then instructed as to correct dietary habits and given the means to do so

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OTHER CASES FOLLOWED UP HAEMATOLOGICALLY AFTER ASCORBIC ACID THERAPY.

No. 167323. Coloured female aged 48 years. Admitted 22nd. March, 1950.

DIAGNOSIS:- Rheumatoid Arthritis with Felty's Syndrome.
3.27 million Red cells per cu. mm.; Haemoglobin 9.0 G.%; Haematocrit 27%; Reticulocytes 0.6%.

No haematological response occurred after 1000 mgm. ascorbic acid daily for one week.

No. 200011. Coloured male aged 20 years.

DIAGNOSIS:- Hodgkin's disease.
Red cells 2.6 million per cu. mm.; Haemoglobin 8.2 G.; Packed cell volume 21.5%.

No response occurred after 1000 mgm. ascorbic acid daily for one week.

No. 195900. Coloured female aged 17 years with rheumatic fever and a packed cell volume of 32.5 %.

No change occurred after vitamin C therapy.

No. 35247. Coloured female aged 33 years with Nephritis with a packed cell volume of 29%.

No response occurred.

No. 156838. Coloured male aged 42 years with aplastic anaemia (Idiopathic) with a packed cell volume of 30% showed no response.

A P P E N D I X III

MEDICINAL PREPARATIONS USED.

FOOD VALUES OF THE CONTROL DIET

THE FOLLOWING PREPARATIONS WERE USED IN THIS INVESTIGATION

1. Ascorbic acid (vitamin C) : "Redoxon" (Roche)

   In most cases in 1000 mg. doses intravenously daily.

2. Vitamin B_{12} : "Rubramin" (Squibb), where 1 ml.=30uG.

   Dose used: 1ml. subcutaneously for 4 days.

3. Pteroyl-glutamic acid (Folic acid) or PGA. :"Folvite" (Squibb). 15 mg. daily for 5 days - orally.

4. Iron :"Ferrivenin"(Benger) where 5 ml. = 100 mg.

   For 5 days 3 to 5 ml. daily was given intra-venously

5. Vitamin B Complex : "Plebex" (Wyeth) or "Bejectal"(Abbott

   1 ml. contains

Thiamine HCl.	10 mg.	11.5mg.
Riboflavin	2 mg.	1.5 mg.
Nicotinamide	100 mg.	57.0 mg.
Pyrodoxine HCl.	5 mg.	-
Ca.Pantothenate	5 mg.	5.7 mg.

   Dose used: 2 ml. by injection daily for 4 or 5 days.

6. Penicillin lozenges (Allenbury's) - one lozenge = 500 units. Given hourly for 3 days.

7. Dihydrostreptomycin (Abbott) ½G. b.d. by injection for 10 days in one case (Case 9)

8. Succinylsulfathiazole : "Sulfa-suxidine" (Sharp and Dohme)

   Used for 5 days (3 G. every four hours) in Case 9.

9. Histamine acid phosphate (B.D.H.)

10. All laboratory reagents used were "Analar".

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FOOD VALUES OF THE CONTROL DIET (Computed as per day)
(FOX AND GOLDBERG (1944).

	Mealie meal	Samp	Bread	Approx. Total per day.
Weight	210 G	50 G	15 ozs.	
Protein	20 G	4.3 G	36 G	54 G
Fat	8.8 G	0.3 G	3.0 G	10 G
Carbo-hydrate	149.1 G	39.4 G	218 G	356 G
Calcium	42 mg.	5.5 mg.	135 mg.	168 mg,
Iron	8 mg	2.0 mg	15 mg	22 mg,
Phosphorus	426 mg	16.5 mg	72 mg.	1021 mg.
Vitamin A	-	-	-	-
Thiamin	0.53 mg	0.03 mg	0.75 mg.	1.13 mg.
Riboflavin	0.25 mg	0.01 mg	0.45 mg.	0.65 mg.
Nicotinic acid	2.52 mg	0.10 mg	13.5 mg	15 mg.
Ascorbic acid	-	-	-	-
Calories	756	177	1035	± 1600

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