A N T E N A T A L G E S T A T I O N A L A G E I N G

By

M. J. BENNETT, M.B.Ch.B.

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A Thesis

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A C K N O W L E D G E M E N T S

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I N D E X

CHAPTER 1

INTRODUCTION

"The duration of pregnancy is not the sole factor determining the biological maturity of a newborn infant but it is the most important" (Farr et al, 1966).

Elective termination of pregnancy plays a small but very important role in current obstetric practice, and as recognition of factors unfavourable to the wellbeing of the fetus improves, this role will increase in magnitude. By elective termination of pregnancy, the author means either induction of labour or elective Caesarean section for any reason whatever. Elective termination of pregnancy in the University of Cape Town obstetric unit accounted for 10.1% of all deliveries in 1968 i.e. 1,313 of 13,034 deliveries (Marais and Strasburg, 1968). In 1969 this figure was 13.8% (Marais and Strasburg, 1969) and in 1970 it was 14.1% (Strasburg et al, 1970).

The obstetrician is often faced with one of two broad problems. The first is the problem of the uterus which is smaller than it is expected to be by dates, when the question of wrong dates or intra-uterine growth retardation must be answered. The second is in dealing with such complications as Rhesus-isoimmunisation, pre-eclamptic toxaemia and diabetes mellitus. The obstetrician is faced with the often very difficult task of timing the termination in such a way as to minimize the risk of prematurity and its attendant hazards, and yet not jeopardize the life of the fetus by leaving it too long in an unfavourable and, as yet, incompletely understood environment. Claireaux (1963) has said, "Prematurity is the most important single factor amongst the first week deaths". The British Perinatal Mortality Survey of 1958 (Butler and Alberman, 1969) found

the commonest denominator amongst deaths following Caesarean section to be prematurity.

From these and other sources, it would appear that the commonest mistake is delivering an infant prematurely as opposed to allowing it to remain too long in the uterus. It is, therefore, vital to have a reliable index of the duration of pregnancy if mistakes are to be minimized.

The usual method of assessing the duration of pregnancy rests in the first instance upon the date of the patients' last menstrual period (L.N.M.P.). It has been found that in Cape Town, 12.5% of all our patients are unable to provide any such date at all, and of the rest, 33% are wrong by more than two weeks when giving the date of their L.N.M.P. Although Lind and Billewicz (1971) claim that patients' dates are unreliable in up to 20% of cases, it would appear that in Cape Town, only a little over half of our patients are able to provide reliable dates. Other methods of assessing the duration of pregnancy are, therefore, required.

The special investigation most commonly employed at the beginning of this study was that of X-ray maturity, but this method is known to yield both false positive and false negative results.

Many methods of estimating the duration of pregnancy have been applied and a voluminous literature bears witness to their inability to solve the problem adequately. The reasons for this appear to be:

i. difficulty in knowing what to measure,

ii. the parameters used may not be a measure of gestational age but possibly some index of biological function which is subject to various influences apart from the passage of time,

iii. nearly all reported studies are cross-sectional rather than longitudinal,

iv. most studies contain various admixtures of babies with growth disturbances which may well interfere with the particular parameter(s) being measured.

It was felt that if an assessment of parameters of gestational age were to be embarked upon, this assessment should be done on normal pregnancies before various pathological states and their possible influences could be considered. With this in view and bearing in mind the magnitude of the problem of the unreliable menstrual histories obtained in Cape Town, it was decided to assess the normal distribution and accuracy of some of the more easily applied parameters of gestational age in a group of normal patients with uncomplicated pregnancies.

Several aspects of this investigation have been studied before. This project as a whole, however, is considered unique in that, to the best of the author's knowledge, no other investigation exists in which so many clinical and biological parameters have been measured, both simultaneously and serially, in normal healthy pregnant patients, and which have been subjected to statistical analysis.

As the majority of clinical problems regarding the duration of pregnancy usually manifest themselves after the 30th week of pregnancy, it was decided to confine the study to this stage of pregnancy.

This study thus comprises easily applied and well recognised parameters, assessed in a control group of patients of known duration of pregnancy, and all studied simultaneously and serially.

The results of the above clinical research form the basis of this thesis.

CHAPTER 2

MATERIALS

Because of the heterogenous population dealt with in our unit and as it was not known whether the race of the patient per se would have any effect upon the data obtained, it was felt that to confine this study to any one particular race group might invalidate the results. Thus it was decided to try to obtain an equal number of European and non-European patients. The author felt that if, upon analysis, any significant racial differences became apparent, correction factors might need to be applied in clinical practice.

As a result of a paucity of information regarding certain of the parameters, especially those of the liquor amnii, in normal pregnancies, the emphasis in this study was based on normality. The data from any patient who, after inclusion in the study group, manifested any departure from accepted clinical norms in pregnancy was immediately discarded, as there was no way of telling what influence this departure had had on any of the data. Similarly the delivery of an infant which was in any way abnormal, whether anatomically or by weight for age definition, precluded the use of that patients' data.

1. PATIENT SELECTION

Patients were asked to participate in the study only if the following criteria were fulfilled:

(a) Gravidity

They were all primigravidae. There were two reasons why it was decided to use primigravidae. Since little or no information is usually available concerning abortions which occurred in the past, it was felt that any patient who had had an abortion, irrespective of the duration of pregnancy, was

best excluded. Secondly, it was felt that the study-patients would probably be required to pay a greater number of visits to the antenatal clinic than they would have done under normal circumstances during the last trimester. The presence of young children at home would have made this exceedingly difficult for multiparous patients.

(b) Age

They were aged between 18 years and 28 years.

(c) Marital Status

A large proportion of the non-European patients delivered in Cape Town are unmarried. This is a result of social customs demanding that a woman prove her fertility before she marries. These women tend to be irregular attenders at the antenatal clinic and generally unreliable and it was, therefore, decided to use only married women. Although not necessarily true in the individual case, as a generalisation married mothers are of higher socio-economic status than unmarried mothers. This is especially pertinent when considering our non-European population.

(d) Rhesus Blood Group

Only Rhesus positive patients were used as it was felt that the risk of producing Rhesus iso-immunisation by repeated amniocentesis in Rhesus negative women could not be justified.

(e) Height

All were of 'average' height i.e. between 150 and 175 cm.

(f) Weight

Their non-pregnant (pre-pregnancy) weight was between 45 and 70 kilograms.

(g) Past Medical History

There must have been no past history of any serious

medical disorders and no patient was accepted if there was an history of any maintenance drug therapy prior to the pregnancy.

(h) Fertility

There must have been no history of involuntary infertility of more than one year's duration.

(i) Past Surgical History

All patients who had had any surgical operation which might conceivably have influenced the course and/or outcome of the pregnancy were excluded.

(j) Progestogens

If progestogens had been used prior to pregnancy, (for any reason including contraception) they were to have been discontinued no less than three calender months prior to the date of the L.N.M.P.

(k) Date of L.N.M.P.

Each patient had to be certain of the date of her L.N.M.P.

(l) Date of Quickening

Each patient had to be able to give the date upon which she first felt fetal movements. The majority of patients were able to do this but a minority were unable to give a precise date, although they could give the week and month during which quickening occurred.

(m) Medication

Patients were allowed anti-emetic preparations early in pregnancy and routine iron therapy only. (All our antenatal patients receive Ferrous Sulphate tabs 2 b.i.d. throughout pregnancy).

(n) Intelligence

Where possible, patients with average or higher intelligence,

as assessed by the author, were selected. It was felt that not only would co-operation be of an higher order but communication between patient and doctor was likely to be easier.

2. CONSENT

Provided all the above criteria had been satisfied, the patient was interviewed and the project fully explained to her. Great care was taken to ensure that the patient understood that the proposed research entailed investigations which were of a non-essential nature, were being done for research purposes and carried slight but definite risk to both mother and fetus over and above the 'normal' risks of pregnancy. The fact that inclusion in the study group was, in fact, a measure of her normality by accepted standards, and that participation was completely voluntary, was stressed. If, after having the proposed study explained, any patient showed signs of hesitancy or fear, she was excluded from the group as it was felt that the possibility of her refusing further participation when half way through her series of investigations was a real one.

Patients who displayed interest were sent home and asked to discuss the matter with their husbands. If there were any objections from the husband, the patient was excluded. If the husband was agreeable that his wife participate in the research project, both husband and wife were interviewed at the next antenatal attendance. At the second visit the entire project was re-explained and any questions answered. If at this point both husband and wife were prepared to give written consent for participation, this was obtained on a specially prepared consent form. (See Appendix A).

3. RESEARCH NUMBERS

Once consent had been obtained, the patient's obstetric

folder was stamped with an easily identifiable stamp and was accorded a research number. The European patients received the uneven numbers viz. 1, 3, 5 etc., and the non-Europeans the even numbers.

4. ANTENATAL VISITS

All the patients were personally seen by the author at each antenatal visit subsequent to their inclusion in the study group. Each patient was seen at two weekly intervals up to the 36th week of pregnancy and weekly intervals thereafter until delivery. At each visit the patients were weighed, blood pressure recordings made, the height of the fundus palpated and the fetal heart auscultated. Urinalysis for the presence of proteinuria and glycosuria was undertaken at each visit. The fundal height and abdominal girth were measured and recorded on a special form. When possible a vaginal examination was made and the state of the cervix assessed. All other information was recorded in the patients' antenatal notes as well as on a special form (Appendix B).

5. LIQUOR AMNII

The intention at the commencement of the study was to obtain specimens of liquor amnii at the 32nd, 35th and 37th weeks of pregnancy. This was not possible in every case because all amniocenteses were done in the Ultrasonic Department and appointments had to be booked to suit both the patients and the Department. Nevertheless a specimen was obtained from the majority of patients within a few days of these dates.

6. PLACENTOGRAPHY AND ULTRASONIC CEPHALOMETRY

Upon the same occasion as each amniocentesis, an ultrasonic examination was made. At the initial visit, placental localisation was undertaken and the result recorded in the patients'

antenatal records. An ultrasonic biparietal diameter measurement was made at each visit to the Ultrasonic Department and recorded upon the patients' research forms, but not on their obstetric records.

7. RADIOLOGY

A single X-ray film of the maternal abdomen was taken on one occasion during the last ten weeks of pregnancy. As it would appear from the literature (Russell, 1969) that the time this investigation yields most information is after the 36th week of pregnancy, an attempt was made to have all patients X-rayed during the 36th week of their pregnancies.

8. LABOUR

Since each patient had developed a certain dependance on him as her doctor, the author offered, whenever possible, to care for each patient during her labour.

Late in the series it was felt that a specimen of liquor obtained in labour would yield additional information and thus an attempt was made to collect uncontaminated specimens by means of a Drew Smythe Catheter once spontaneous labour had begun. This was not always successful as there was often either insufficient liquor or the forewaters were accidentally ruptured rendering the liquor contaminated by maternal secretions. Thus in only a minority of patients were four specimens of liquor obtained.

9. PUERPERIUM

Following delivery the mother and infant were managed routinely unless circumstances dictated otherwise.

(i) Mother: No mother having had her first baby is discharged from our unit until she has been shown how to bath, dress and feed her baby. If non-absorbable suture material has been

used for the suturing of episiotomies, perineal tears etc., she remains in hospital until these are removed on the fifth day after delivery. All are given an appointment to return six weeks later for a postnatal check-up.

(ii) Infant: Each infant born in our unit is seen and examined by the resident Paediatric Registrar, and the infants born of the mothers under investigation were no exception. One Paediatric Consultant at each of the two maternity hospitals concerned saw each of these infants within 48 hours of delivery and assessed its gestational age. This was recorded upon both the infants' notes and the mothers' study form. (Appendix B).

(iii) Postnatal Visit: Each mother was asked to return for a postnatal check-up approximately six weeks after delivery and to bring her baby with her. At this visit the mother underwent a full general and gynaecological examination and a Papanicolaou smear was taken. Each infant was weighed and fully examined and the results of these examinations recorded in the respective sets of notes. Provided all was normal, as was the case with each of the study patients, the question of contraception and family planning was discussed with each patient prior to her discharge.

CHAPTER 3

METHODS

A brief account of the methods used in this investigation now follows. Where well documented techniques were used only the principle is outlined, but where techniques were modified for the purposes of this investigation, a detailed account is given.

It must be stressed that at no stage during the course of the investigation was any attempt made to interpret the results obtained from any of the parameters studied, as it was felt that this would invalidate the value of these and possibly future results.

The author alone knew how each research number was allocated and as little information as possible concerning individual patients was passed on to colleagues partly involved in the programme. This meant that, for example, the doctor making ultrasonic biparietal diameter measurements was not told the duration of pregnancy of any of the patients involved, and could thus not be biased by what he would have expected a measurement to be, had he known the duration of pregnancy. Knowledge of results of previous and other investigations was similarly withheld in an attempt to exclude bias.

1. CLINICAL EXAMINATION OF THE FUNDAL HEIGHT

The traditional relationship of the uterine fundus to abdominal landmarks at different gestational intervals throughout pregnancy was used. (The Queen Charlotte's Textbook of Obstetrics, 1970).

2. FUNDAL HEIGHT IN CENTIMETRES

This was measured with the patient lying supine. A standard metric tape was used and a measurement in centimetres was

made of the height of the uterine fundus above the upper border of the symphysis pubis. This measurement was made along the long axis of the uterus and after the uterus had been centred in the abdomen. (McDonald, 1906).

3. ABDOMINAL GIRTH

This was measured with the same tape with the patient again supine. The measurement was taken through what was estimated to be the plane at right angles to the long axis of the uterus. In this plane the maximal girth measurement was recorded.

4. CALCULATED UTERINE VOLUME

Using the measurements obtained in 2. and 3. above, the uterine volume was calculated using a conversion of the formula for the calculation of the volume of an ellipse, namely:-

$$\text{Volume} = {}^{4}/_{3}\pi\, r^{1}\, r^{2}\, r^{3},$$ assuming that rotation about the long axis is symmetrical.

The formula used was:

$$\text{Volume} = {}^{4}/_{3}\pi \left(\frac{G}{2\pi}\right)^{2} \times \frac{F.H.}{2 \text{ or } 3}$$

where G = Abdominal Girth in centimetres

and F.H. = Fundal Height in centimetres

In view of the fact that this calculation is inaccurate, two allowances had to be made in order that its accuracy was not further undermined by normal characteristics of advancing pregnancy.

Firstly, as this division of the fundal height by 2 produces only an approximation of the radius of the long axis, it is obvious that when dealing with a markedly convex abdomen, this

division is insufficient. To correct the error arising under this latter circumstance, the divisor was increased to 3 i.e. $\frac{F.H.}{2}$ was used unless the abdomen was markedly convex forwards when $\frac{F.H.}{3}$ was used. The choice of division was not based upon any scientific measurement but on the author's opinion concerning the protruberance or otherwise of the abdomen.

Secondly, the measurement of F.H. is more correct when the fetal head is unengaged in the maternal pelvis and theoretically virtually all of the uterus is in the abdomen. When the fetal head has become engaged in the maternal pelvis a proportion of the uterine contents, and thus volume, is not included when the measurement is started at the symphysis pubis. To allow for this, 5 centimetres are added to the fundal height reading when engagement of the fetal head has occurred. When the head was found in an intermediary position in this study, an attempt at correction was made by adding fractions of 5 to the fundal height reading, depending upon the amount of head immeasurable, as assessed clinically.

Neither the basic measurements, corrected or uncorrected, of fundal height or abdominal girth, or the calculated uterine volume were entered in the patients' obstetric notes.

5. MATERNAL WEIGHT

Each patient was weighed at each visit. To avoid the error introduced by the variations in weights of items of clothing, each patient was weighed in her underwear only. All weight readings were obtained on the same scales; one in each of the two hospitals. Conversion from the Avoirdupois to the Metric systems was achieved by means of conversion tables. (Documenta Geigy, Scientific Tables, 6th Ed.)

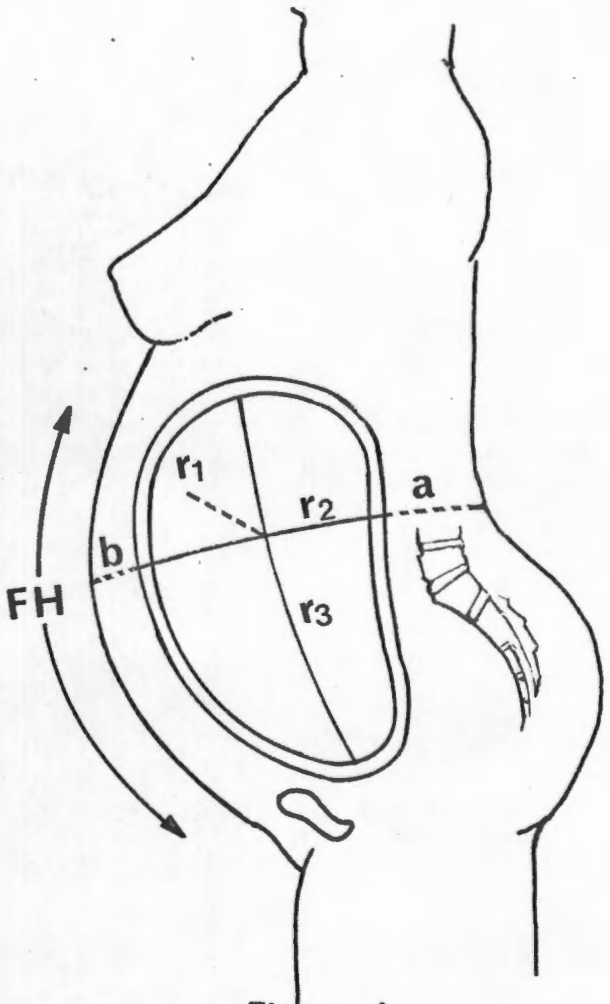


Figure 1.

UTERINE VOLUME = $\frac{4}{3}\pi r_1 r_2 r_3$

$$\therefore \frac{4}{3}\pi \frac{G}{2\pi} \frac{G}{2\pi} \frac{F.H.}{2 \text{ or } 3} = \frac{4}{3}\pi \left(\frac{G}{2\pi}\right)^2 \times \frac{F.H.}{2 \text{ or } 3}$$

(assuming symmetrical revolution)

Diagramatic representation of the derivation of the formula for measuring uterine volume.

6. ASSESSMENT OF THE CERVICAL STATE

At each visit after 30 weeks, a vaginal examination was performed and the state of the cervix assessed. Three cervical characteristics were assessed viz.

i. Position of the cervix

ii. Effacement of the cervix

iii. Dilatation of the cervix

A system of numerical grading of the cervix was drawn up with the ultimate aim of statistical analysis of this parameter. This system was as follows:

Grade 1: neither effaced nor dilated enough to admit one finger (1F)

Grade 2: either effaced or dilated enough to admit 1F

Grade 3: both effaced and dilated enough to admit 1+F but posteriorly situated. (1+F corresponds to a diameter of two or more centimetres).

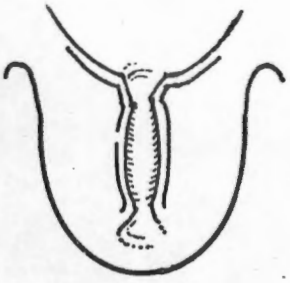
Grade 4: the same physical characteristics as for Grade 3 but now centrally situated.

In assessing the dilatation, no forceful attempt was made to introduce the finger into or through the cervical canal as it was felt that this would cause pain to the patient and might prejudice the onset of spontaneous labour. Although strict asepsis was not observed during this procedure, sterile rubber gloves were worn.

7. AMNIOCENTESIS

This was done as a sterile procedure. It was felt that the risk of infection if the procedure were done in the antenatal clinic was greater than if a less frequently used room were utilised. Because ultrasonic cephalometry was to be done at the same periods of gestation, it was felt that the Ultrasonic Department was the best

GRADE 1.



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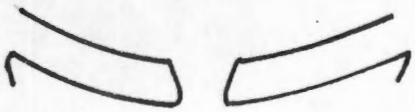

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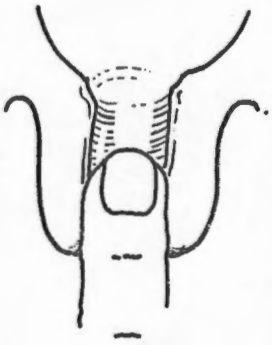

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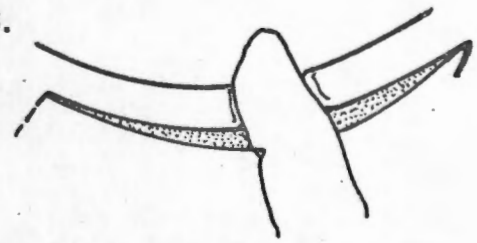


FIGURE 2.

Diagram to show the changes assessed in the grading of the cervical state.

place for amniocentesis to be performed.

The technique was explained to each patient prior to the initial tap and she was warned about what to expect. Prior to the first amniocentesis the placental site was localised and ultrasonic cephalometry performed. (See section 12.)

Maternal venous blood was taken and placed into a glass test-tube. One drop of blood was evenly spread on a clean glass slide and allowed to air dry for later evaluation by means of the acid elution technique (see section 11.). The patient was then asked to empty her bladder following which the fetal heart was auscultated.

With the author fully scrubbed, gowned, gloved and masked, the patient's abdomen was cleaned with an antiseptic solution (0.5% Hibitane in 70% Alcohol) and draped with sterile towels. The site of proposed puncture was decided upon after careful abdominal palpation.

At the 32nd week of pregnancy, the fetal head could easily be moved in a cephalad direction and a gap thus created between it and the pubic symphysis (see Figure 4). Local anaesthetic (1% Lignocaine) was slowly injected into the skin and abdominal wall down to the peritoneum at the site chosen for puncture. To avoid possible trauma to blood vessels this was in the midline if possible. Injection of local anaesthetic solution into the myometrium was avoided as this has been reported to cause areas of myometrial activity and even uterine contractions, rendering the procedure more difficult and possibly more dangerous (Gordon, 1969).

With the left hand holding the fetal head away from the puncture site, the 12.5 centimetre 20 gauge needle with obturator was firmly introduced into the uterine cavity. If, upon withdrawal of the obturator, a free flow of liquor amnii was obtained, a sterile syringe was used to aspirate approximately 20ml of amniotic fluid. The needle

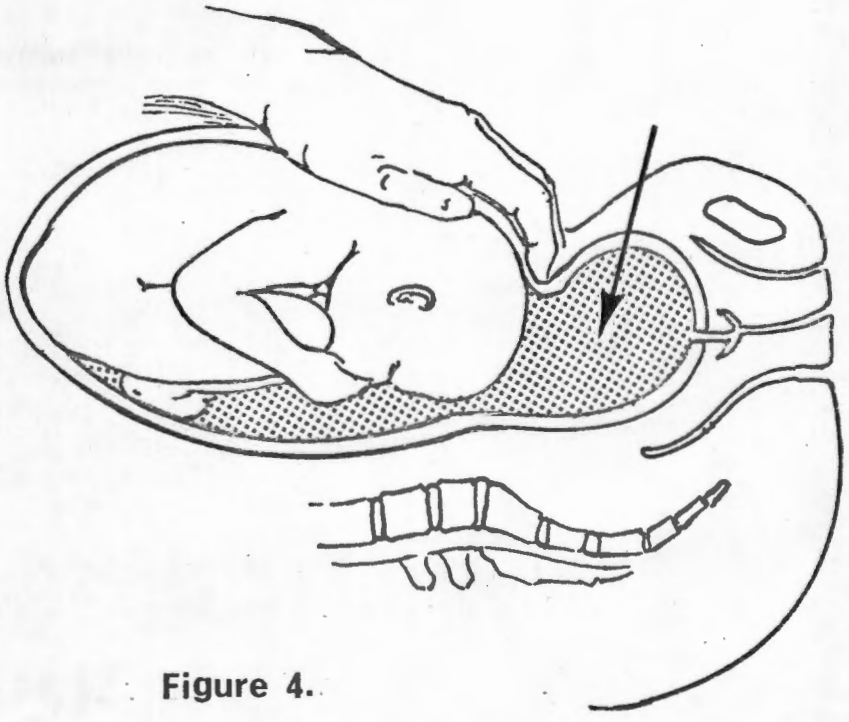


Figure 4.

A lateral view of the pregnant uterus showing the infra-cephalic site of puncture for amniocenthesis.

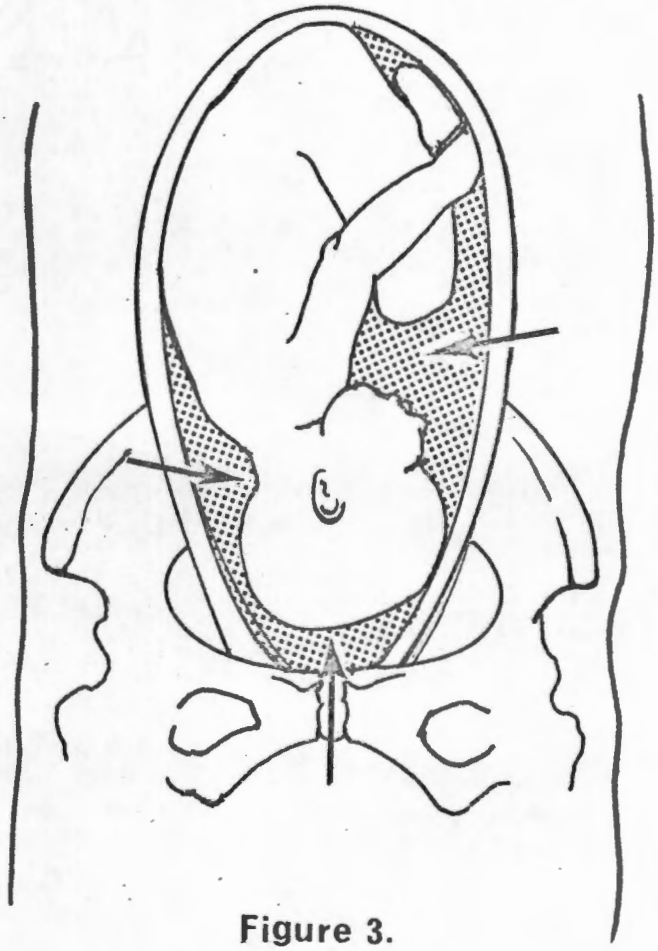


Figure 3.

An antero-posterior view showing the sites of puncture for amniocenthesis

was then swiftly withdrawn, the puncture site sprayed with Dermoplast* and a small sterile gauze dressing applied.

The liquor was immediately placed into two 10ml test-tubes and one of them placed in a light-proof envelope. The fetal heart was again auscultated. A drop of maternal capillary blood was obtained from a finger prick and spread thinly and evenly on a clean dry glass slide and allowed to air dry. The tubes of blood and liquor amnii and the two smears were marked with the patients research number and the date, and all were then taken to the laboratory.

Sometimes at the 35th week and nearly always at the 37th week of pregnancy, the fetal head was found to be engaged in the maternal pelvis. This meant that the technique already described for amniocentesis could not be used. One of two alternative methods was then used depending upon the location of the placenta. If the placenta was posteriorly situated, then the area between the fetal trunk, upper and lower limbs was used as the site of puncture. When the placenta lay anteriorly or antero-laterally, the site for puncture lay over the nape of the fetal neck i.e. liquor was obtained from the space between the fetal occiput and back (Figures 3 and 4).

8. LIQUOR CYTOLOGY

In the majority of cases the liquor was examined within one hour of amniocentesis. The liquor was initially examined macroscopically and described in the following way:

i. colour
ii. presence or absence of turbidity
iii. presence or absence of blood
iv. presence or absence of meconium
v. presence or absence of a 'surface layer of cells' in the supernatant after centrifuging.

*Ayerst Laboratories Incorporated

Prior to centrifuging, 1ml of liquor was removed for staining with the Nile Blue method. Thereafter the liquor was spun at 2,000 r.p.m. for three minutes and the supernatant decanted for bio-chemical analysis. The deposit was then spread evenly on a clean glass slide and whilst still wet, fixed with a spray fixative containing Polyethylene Glycol and Alcohol.*

A. Fat cell count using Nile Blue Sulphate stain

Nile Blue Sulphate solution was made according to the method of Pearce (1960). The method is as follows:

Nile Blue**	1 gram
Pure Sulphuric Acid	0.5ml
Water	100ml

This was boiled for two hours with occasional additions of water. It was then allowed to cool and was then filtered through a standard Watmans No. 45 paper. The volume was then made up to 100ml with water to produce a 1.0% solution. A 1 in 10 dilution was then made of 10ml of this solution to produce a 0.1% solution.

Using the method described by Brosens and Gordon (1965), it was found that drying of the slide occurred rapidly and when this occurred the slide was unsuitable for evaluation. In view of this, a modification of the Nile Blue Sulphate staining method was evolved. This modification was used throughout this study once it had been found to produce a stable stain which allowed accurate evaluation of the cells.

The modification was as follows: two small test-tubes were each instilled with an equal volume of liquor amnii

* Fencott; Sangene Products (Pty) Ltd.

** Supplied by George T. Gurr Ltd., London.

(uncentrifuged). Into the first tube, an equal volume of 0.1% Nile Blue Sulphate was placed. Into the second tube a similar volume of 1.0% Nile Blue Sulphate was added. The volumes of liquor amnii and Nile Blue Sulphate were 0.5ml in each instance. Both tubes were then placed in a water-bath at 37°C for five minutes with occasional gentle shaking to ensure adequate mixing. A drop of each solution was then placed on a clean glass slide and covered with a coverslip. The drops were placed at opposite ends of the same slide to allow for easy comparison.

A total of 300 cells was counted and the 'fat cells' expressed as a percentage of the total. The presence or absence of clumping of fat cells was noted and an attempt made to grade this into the categories of none, minimal, moderate and diffuse.

Although 0.1% Nile Blue Sulphate proved to be the better of the two concentrations for staining purposes in the vast majority of instances, occasions arose when the 1.0% solution provided the better picture.

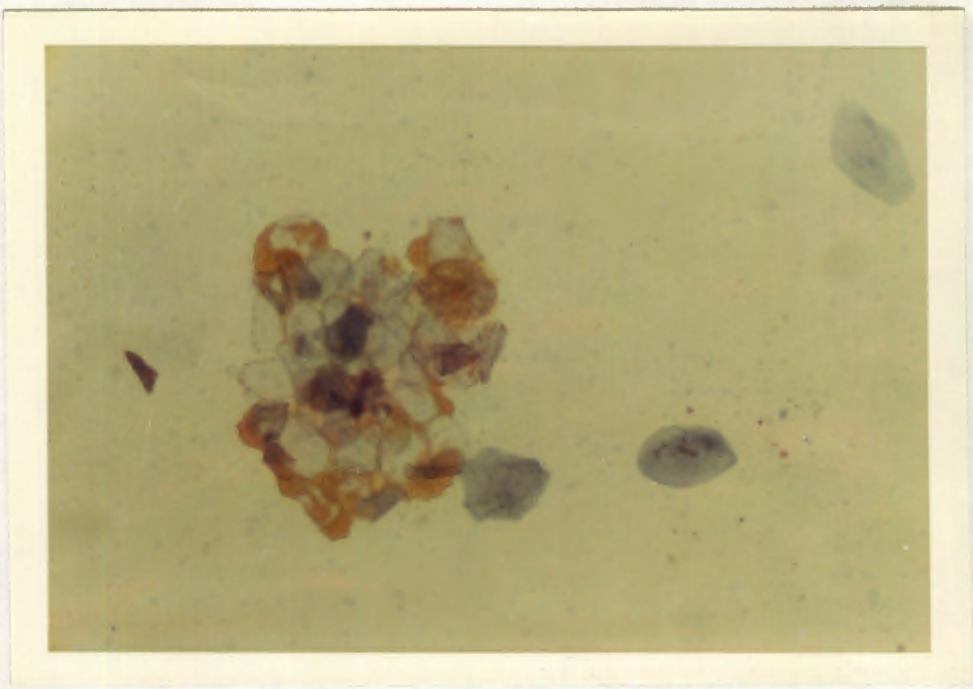
B. Differential cell count using the Harris-Shorr stain

This stain was selected because it provides a sharp differentiation between cornified and non-cornified elements and was adopted unmodified from Drury and Wallington (1967). The slide with the cellular deposit (see section 8.) was stained by this method and a differential cell count was then done using morphological criteria similar to those used in vaginal cytology.

The types of cell differentiated were:

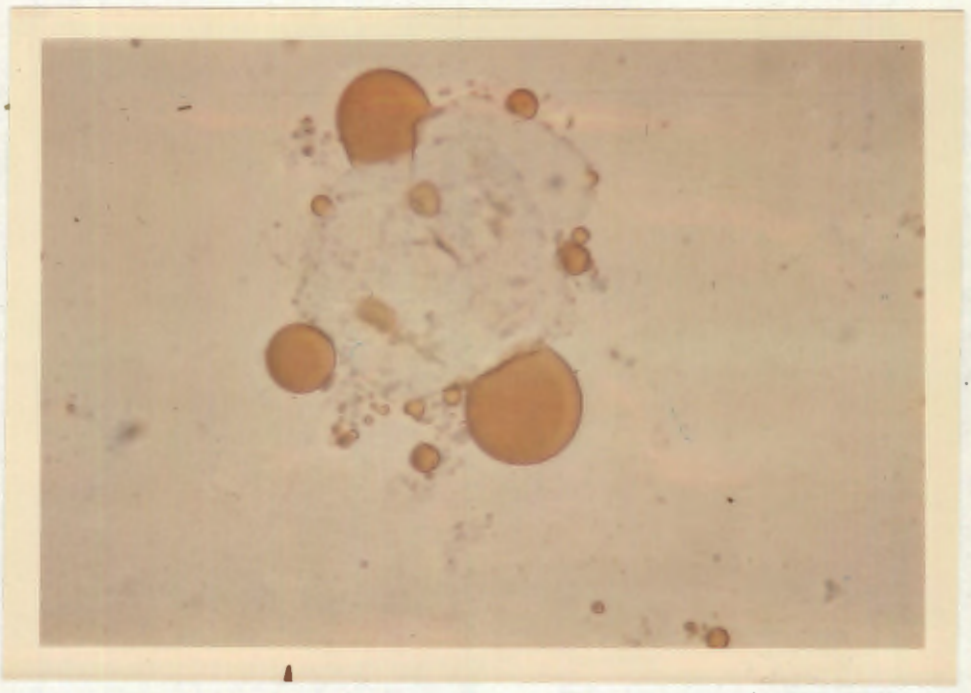
i. Polygonal cell: the outline approximates a regular pentagon or heptagon. The cytoplasm is transparent with a filligree or granular aspect. Although the nucleus is always absent, there is

Figure 5



Typical clumping of fat cells stained with Nile Blue (x 200)

Figure 6



A fat cell stained with Nile Blue, showing the typical extra-cellular fat globules (x 400).

often an annular structure visible in the centre of the cell, probably indicating the former nuclear site.

ii. Anucleate Squame: large polygonal-shaped and with no discernable nucleus.

iii. Superficial Squame: large, flat polygonal-shaped cells with a pyknotic structureless nucleus of less than 6 microns in diameter.

iv. Intermediate Squame: large, flat polygonal or slightly rounded cell with a vesicular nucleus greater than 6 microns in diameter.

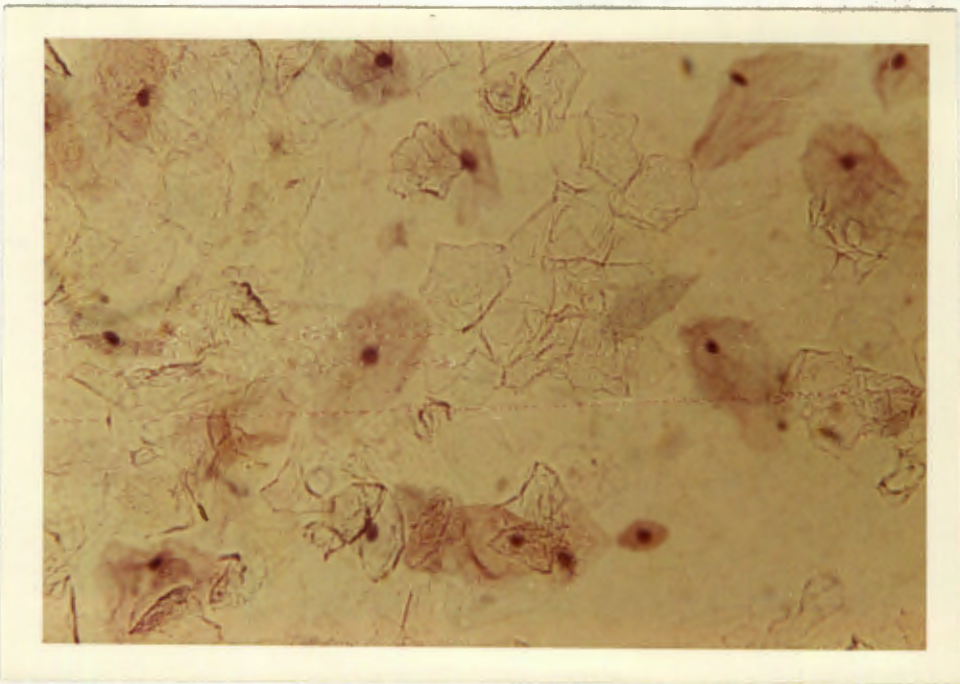
v. Basal Cell: smaller than the above cell types and round in shape. The nucleus is less vesicular than that of the intermediate cell, is usually concentrically situated, and the nuclear-cytoplasmic ratio is greater.

vi. Amnion Cell: variable in shape from round to globular with a dense and often vacuolated cytoplasm. The nucleus is usually eccentrically situated. The amnion cell is smaller than the basal cell.

The staining characteristics of the cells described have been well documented by Huisjes (1970) and, as this count was based upon morphological criteria, play no role at this point. They will, however, be discussed briefly at a later stage.

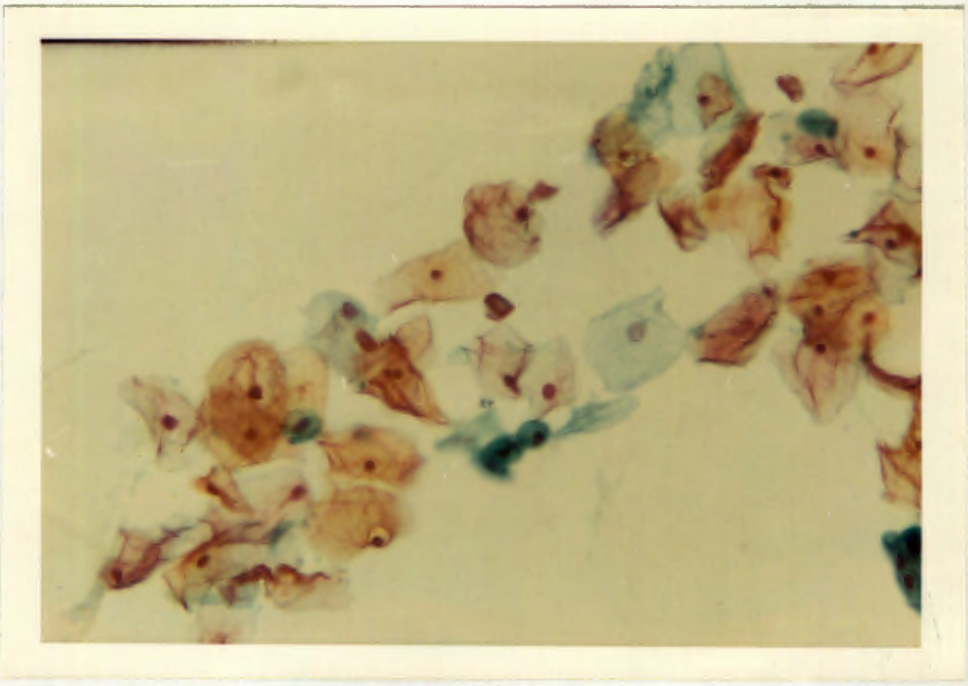
A total of 500 cells was counted on each slide and the

Figure 7



Typical translucent polygonal cells stained with the Harris-Shorr stain (x 200)

Figure 8



An example of the colour range and low power detail of typical cells found in liquor amnii stained by the Harris-Shorr method (x 200)

G E S T A T I O N A L A G E I N G R E S E A R C H

LIQUOR AMNII CYTOLOGY

PATIENT NO: DATE SPECIMEN OBTAINED:

1. NAKED EYE DESCRIPTION .....

.....

2.	MACROSCOPIC BLOOD		MICROSCOPIC BLOOD		NO BLOOD	

3. NILE BLUE

Clumping	Nil	Minimal	Moderate	Diffuse

	Number	%
Fat Cells		
Blue Squames		
Yellow Anucleates		

4. HARRIS-SHORR

	Number	%
Polygonal Cells		
Anucleate Squames		
Superficial Squames		
Intermediate Squames		
Basal Cells		
Amnion Cells		

5. 'KLEIHAUER'

Pre-Amniocentesis

Post-Amniocentesis

FIGURE 9: An example of the form used for recording the results of cytological examination of liquor amnii

proportions of each cell type expressed as a percentage of the total.

Results of the cytological examination of liquor amnii were recorded on the form shown on page 26 and no attempt was made to interpret them. None of the above results were recorded in the patients' obstetric notes.

9. BIOCHEMISTRY

In spite of the fact that autoanalyser facilities are available in the Chemical Pathology Department at Groote Schuur Hospital and are extensively used in the routine services provided by this Department, it was decided that for the purposes of this study a laboratory would be set up with the sole purpose of doing the bio-chemical estimations required. This laboratory was set up in the Department of Obstetrics and Gynaecology at the Medical School and was placed under the supervision and control of the author. It was felt that bio-chemical estimations done manually, although losing the reproducibility achieved by the autoanalyser process, would achieve a higher degree of accuracy, provided they were all performed under the same conditions. The laboratory was staffed by a qualified medical technician who was employed on a part-time basis and with whom the author worked in close collaboration.

In view of the volume of work entailed by the project as a whole, all the bio-chemical estimations were carried out by the medical technician once the author had assured himself that each of the chosen methods gave reproducible and accurate results and could, if warranted, be used without modification as part of a gestational ageing service.

Initially an attempt was made to perform all bio-chemical estimations on both the liquor amnii and the maternal serum on the

same day as they were collected. It very soon became apparent that this was not always possible as the laboratory functioned in the mornings only and some amniocenteses were done on Wednesday afternoons. To circumvent this problem the samples obtained either too late in the morning for analysis that day or those collected in the afternoons were immediately placed in a refrigerator at -20^{o}C for analysis the following day.

This step was only taken when the effects of freezing upon the bio-chemical constituents of liquor and serum had been studied. Samples of both liquor and serum were divided equally; the first fraction was examined bio-chemically immediately, whilst the second fraction was frozen at -20^{o}C. The following day i.e. $\pm$ 24 hours later, the second fraction was unfrozen and the same bio-chemical tests were then performed. The results of the tests were then compared to assess the effect of freezing. This experiment was repeated upon different samples of both liquids with different concentrations of the chemical substances being estimated. The results of this experiment showed that although differences in the levels of chemicals estimated did occur, they did so in a completely random fashion and they were within the ranges of experimental error for the methods used (Appendix C). Thus it was concluded that the effect of freezing the specimens over a period of 24 hours or less could be ignored for the purpose of this research project.

Although each method used was checked for accuracy and reproducibility within an acceptable range of experimental error for that method, prior to the commencement of the project (Appendix C), a standard control solution was prepared and run with every batch of estimations for each of the chemicals determined. This standard solution varied in its concentration in order to cover the range of

results obtained from the actual estimations.

The same methods were used for both the liquor amnii and the serum estimations and thus only a single account is given of the methods used.

A Unicam colorimeter incorporating an A-filter was used for the colorimetry. This filter has a transmission of 490 millimicrons.

a. Urea

The concentration of urea in maternal serum and liquor amnii was determined by the Urease Nesslerization Method. This method utilises the conversion of urea to ammonium sulphate by the action of the enzyme urease. The ammonia formed is estimated by Nesslerization. (Varley, 1962a).

b. True Creatinine

The method for the determination of true creatinine was that of Owen et al (1954) making use of an hydrated aluminium silicate (Lloyd's reagent) to adsorb the creatinine fraction and thus allow separation from the other 'chromogens' before determination.

c. Creatinine

For the determination of creatinine, the modified Folin-Wu Tungstic Acid method was used for the precipitation of the proteins followed by the Jaffe Picric Acid method of the production of a red colour with an alkaline picrate solution. (Varley, 1962b).

d. Bilirubin

The method for the determination of bilirubin was that of Michaëlsson (1961). This is basically a diazo method which makes

use of the reaction between bilirubin and diazobenzene-p-sulphonic acid to form a coloured coupling product, an azo pigment. The Michaëlsson method is a modification of the method of Jendrossik-Gróf-Nosslin and was chosen because it eliminates the error in bilirubin estimation caused by the products of haemolysis i.e. haemoglobin. In this modification, ascorbic acid is added to the total bilirubin sample after completed diazo-coupling.

10. RADIOGRAPHIC TECHNIQUE

A conscientious attempt was made to have abdominal X-rays taken of each patient during the 36th week of her pregnancy. At the time of this investigation, the accepted teaching was that the distal femoral epiphyses appeared radiologically at 36 weeks. As this is the accepted arbitrary point delineating prematurity from maturity in our unit, it was felt that the 36th week of pregnancy was an ideal time to assess 'fetal maturity' radiologically, as well as providing an index of the development of the distal femoral epiphysis in the patients under the care of our unit.

The non-White patients in the study group were all delivered at Groote Schuur Hospital and were X-rayed there. Two different techniques were used for taking the X-ray pictures. All the patients at Groote Schuur Hospital are X-rayed in the true lateral position, whereas all patients X-rayed at Mowbray Maternity Hospital have a postero-anterior film taken.

a. Radiological Apparatus

The same apparatus was used at both hospitals.

i. Type of Unit: a fully rectified six valve Siemens unit with rotating anode tube.

ii. Type of Film: Kodak Rapid Processing films

iii. Size of Cassette: 35cms x 42cms for both the lateral and postero-anterior films.
25cms x 20cms for the 'coned' views (vide infra)

iv. Processors: Kodak X-omat 90 second processors were used. The films are developed in rapid chemicals and high temperatures. Automatic dryers were used to complete this processing.

v. Type of Screen: Phillips Universal screens

b. True Lateral Technique

Following micturition the patient is placed prone on the X-ray table in either the left or right lateral position. The maternal spine is removed from the centre of the table i.e. the lateral aspect of the abdomen is over the centre of the table. A 25cms long and 10cms wide bag, firmly filled with cotton wool, is placed immediately anterior to the patient's spine underneath the patient, extending from approximately the fourth lumbar vertebra to the xiphisternum. A second such pad is placed on the opposite side of the patient's abdomen and a compression band applied. This has the effect of pushing the uterus forwards and thus clearing the fetus of the maternal spine. The position of the patient is checked and the true lateral position verified. A pillow is placed between the patient's legs (from knee to ankle) which are extended and her arms are brought up near her head.

The anode is centred at the level of the iliac crest and midway between the maternal spine and the anterior abdominal wall.

Tube-film distance	:	100 cms
Kilovoltage Peaks (K.V.P.).	:	75 - 95
Milliampere-seconds	:	80
Duration of exposure	:	less than 0.5 seconds

With this technique, a single exposure is all that is required.

c. Postero-anterior technique

With the patient in the prone position, suitably supported on pillows under her chest, hips and ankles, the film is centred to include the lower border of the symphysis pubis. The centre point of the ray is marked on the patient's skin. The film is exposed with the patient holding her breath after complete expiration.

After exposure of the film, the patient is requested to remain quite still and the film is processed as rapidly as possible.

If the fetal knee joints are clearly demonstrated, no further film is taken. If, however, this is not the case, the position of the knees on the X-ray film is carefully noted and, if necessary, measured, and making use of the skin mark indicating the centre of the first film, the anode tube is centred in a manner calculated to give a localised view of the knees. It has been found that when necessary, it is sufficient to rotate the patient through approximately 10^{o} one way or the other in order to throw the fetal knees clear of the maternal spine.

Should a second film be necessary, a close colimater for the 25 x 20cms film is used. When an oblique view is decided upon, the kilovoltage is increased by 10 - 20 K.V.P. to accommodate the increase in tissue mass. As described above a second exposure is sometimes necessary.

d. Radiation Dosage

The theory that irradiation in low dosage may be a cause of fetal abnormality or disease cannot be sustained with absolute certainty in humans because of the many variables, the absence of precise dosage and the fact that no prospective trials, in sufficient numbers, have yet been done. Nevertheless, Stewart et al (1958), in a long term follow up at Oxford, concluded that there was a 40% increased risk that leukaemia and other childhood neoplasms may follow irradiation in utero. Gruin et al (1967) found a non-significant incidence of leukaemia, neurological and ophthalmic changes but a significant increase in the incidence of haemangiomata.

When considering the levels of irradiation at which damaging effects might be expected, Rugh (1968) postulates the changes as follows:

> "During the pre-implantation (0-9 days) irradiation tends to cause death rather than anomalies: during the phase of active organogenesis (9-42 days) it can cause severe structural anomalies, at diagnostic levels of exposure of 25 rads (R) or more; and during the succeeding weeks of pregnancy the resulting amonalies tend to be functional rather than structural and may be obvious, even in the neonatal period."

With the techniques used in this study, the calculated maximum fetal dose was 1 rad, even when two exposures were used, (Werbeloff, 1971). Assuming that the hazard is proportional to the X-ray dose received by the fetus, and remembering that no fetus in this study was irradiated prior to the 35th week of pregnancy, the author felt that a maximum dose of 1 rad at this

stage of pregnancy was, in the light of present knowledge, a safe dosage for the purposes of this study.

e. The radiological method of estimating gestational age

Every standard textbook on obstetrics lists the time in pregnancy when particular skeletal ossification centres become radiologically evident. These have long been used as a guide to the prediction of fetal maturity as advocated by Hartley (1957). Although many factors have been reported to play a role in the development of osseous centres e.g. race and sex of fetus (Christie, 1949), 'intra-uterine malnutrition' (Usher, 1970), diabetes mellitus (Russell and Rangecroft, 1969) and congenital malformations such as spina bifida and anencephaly (Russell and Rangecroft, 1969), many authorities accept that in the absence of pathological states, their appearance on X-rays correlates well with the duration of pregnancy (Greenhill, 1961).

For the purposes of this study and because 39 of the 47 patients (83.0%) were X-rayed after the start of the 36th week, only two of the many centres were examined. As shown by Hartley (1957) the presence of the distal femoral epiphysis was taken to indicate a maturity of 36 weeks and the presence of the proximal tibial epiphysis as indicating 38 weeks.

Such things as fetal size and the length of various long bones were not taken into account as it was felt that they were liable to both subjective and technical errors.

11. ACID ELUTION OF BLOOD SMEARS

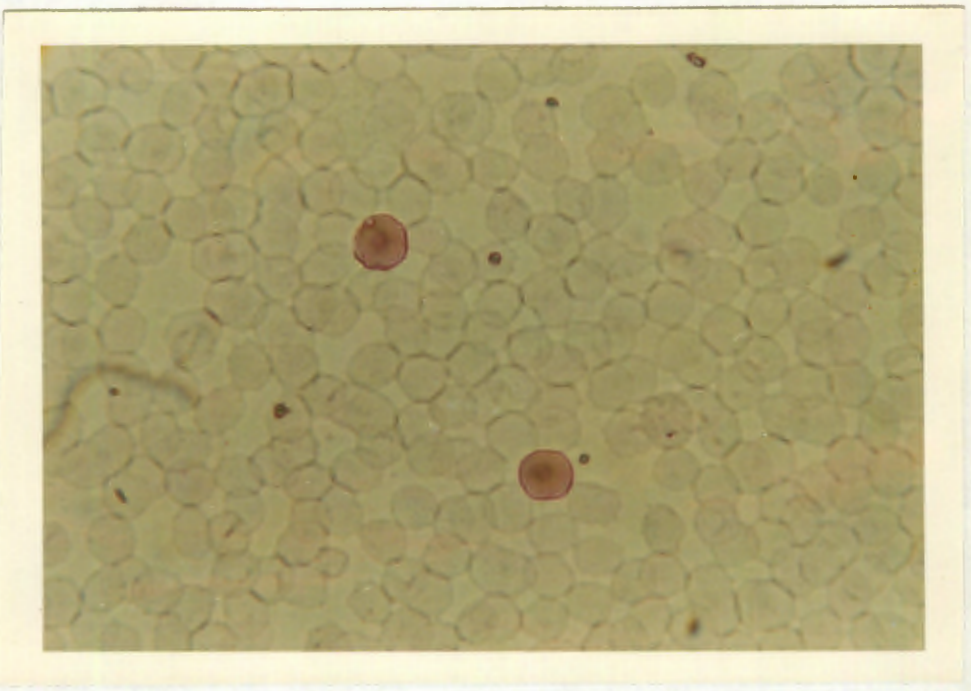
Zipursky, Pollock, Chown and Israel (1963) showed that with amniocentesis, not only was a large trans-placental haemorrhage demonstrable in some instances, but in these cases a sudden sharp rise

in antibody titre followed where the fetus was Rhesus positive and the mother Rhesus negative.

This test was performed in order to know the magnitude of the risk of producing a feto-maternal micro-transfusion by the procedure of amniocentesis. As all the patients in this study group were Rhesus positive primigravidae, the possibility of iso-immunisation being either caused or worsened by this procedure was eliminated. Nevertheless, as it was hoped at the start of this project that some of the parameters to be assessed would prove to be of value in the estimation of gestational age, and as it was forseeable that either unsensitized or already sensitized Rhesus negative patients would require such investigation for clinical reasons, it was deemed necessary to evaluate the possible risks attendant upon any of the techniques used.

Maternal venous blood was taken prior to every amniocentesis and a drop of capillary blood obtained by finger prick was collected approximately twenty minutes after each amniocentesis. Pre- and post-amniocentesis blood smears were made by thinly spreading a drop of blood on a clean glass slide and allowing it to dry in air at room temperature. These blood films were stained by the Liverpool modification of the Kleihauer technique (Woodrow and Finn, 1966). This stain makes use of the fact that by treating fixed blood smears with a buffer of Citrate-Phosphate solution of pH 3.3 at 37^{o} Centigrade for two minutes, adult haemoglobin (Hb-A) can be eluted from erythrocytes while fetal haemoglobin (Hb-F) remains in the cells. The pH of the buffer is critical in that complete elution of Hb-A does not occur consistently at pH 3.4, whereas it does at pH 3.3. Staining was by means of Ehrlichs Haemotoxylin for one minute and counterstaining in 2.5% Aqueous Eosin for two minutes. Controls,

Figure 10



A high power picture of two fetal erythrocytes in a field of ghosted maternal erythrocytes obtained by means of the acid elution technique.

both negative and positive, were frequently used. Negative controls were made from smears of umbilical cord blood diluted 1:1,000 adult blood.

The smears were examined microscopically and the number of fetal erythrocytes expressed as the number of fetal cells in 50 square millimetres (mm^2) of blood film. The area comprising one low-power field is calculated from the formula πr^2.

On the Leitz Vetzlar microscope used by the author, the low-power lens yielded a field with a diameter of 1.36mm and a radius of 0.68mm, giving an area of $1.453mm^2$. To obtain an area of $50mm^2$, 34.4 low-power fields had, therefore, to be examined. As the fraction could not be assessed, 35 low-power fields were examined i.e. $50.9mm^2$.

A count of 5 cells per 50 low-power fields has been calculated to represent 0.25ml of fetal blood in the maternal circulation (Combined Study, 1966). As only 35 low-power fields were being examined it was felt that a figure of 3.5 cells per 35 low-power fields could be accepted as being an approximately equal concentration and thus a rough guide as to the size of a feto-maternal transfusion, when thus diagnosed, could be obtained.

12. ULTRASONIC PRINCIPLES

Since September 1970, ultrasonics has played an increasingly important role in obstetric practice in the Cape Town unit. The apparatus installed at that time was the diasonograph (Nuclear Enterprises Limited) and is at the time of writing, one of the only two such pieces of equipment in the country.

The diasonograph probe contains a piezo-electric crystal of barium titanate and there is a choice of three probes, each producing ultrasonic energy waves of differing frequency. The three

frequencies available are 1½ megacycles per second (1.5MHz), 2½ megacycles per second (2.5MHz) and 5 megacycles per second (5.0MHz). For general purposes the 2.5MHz probe is used, but when greater penetration is required, as for example in obese patients, the 1.5MHz probe is used.

Ultrasound is pulsed at a repetition rate of 300/second and the principle is that at the frequencies used for diagnostic work, it can be propagated under directional control as a beam. Partial reflection of the beam occurs when it crosses a boundary or interface between tissues of differing physical properties and predictably so if the interface is encountered at perpendicular incidence. Ultrasound is believed to obey the same laws of reflection and refraction as light.

The electric signals arriving at the piezo-electric crystal are converted into pulsed ultrasonic waves of the chosen frequency. The ultrasonic beam so formed is directed into the tissues to be examined. The amount of energy reflected depends upon the difference in the physical properties of tissues bounding the interfaces encountered. The residual energy continues inwards into the depths of the tissues until the next interface is encountered and so on, the beam undergoing progressive attenuation. The echoes return to the piezo-electric crystal in the transducer mounting and are converted into electrical signals which are amplified and handled by a cathode-ray oscilloscope. The same piezo-electric crystal is able to convert electric impulses into ultrasound and then convert the returning echoes back into electrical signals because, although it is emitting ultrasonic pulses at a rate of 300/second, these are of extremely short duration and for the major portion of the time it is quiescent and acting as its own echo receiver.

As air does not transmit ultrasound, the method of direct contact scanning (Donald, 1968) is used in Cape Town. The probe is applied directly to the skin of the abdominal wall, making use of a film of olive oil to secure acoustic coupling. This method produces only minimal discomfort to the patient.

a. Ultrasonic Display Systems

Two different oscilloscope display systems are available and both were used in this study.

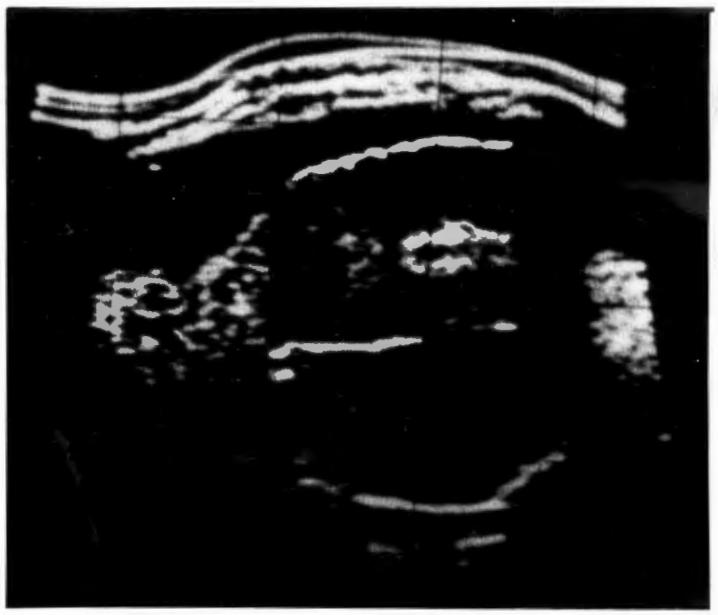
The A-ecan is a unidimensional display wherein the returning echo signals are applied to the deflecting plates of the cathode ray tube so that the point of light forming the time-base sweep across the tube is deflected upwards as a peak whose height corresponds to the strength of the echo, and whose position from left to right across the tube corresponds to the depth of its point of origin. As the distances between the leading edges of these peaks can be accurately measured electronically, this technique is useful in measuring a diameter whose existence and position are already known. (Donald, 1968).

The B-ecan system is two dimensional. The returning electrical signal is applied to the grid of the cathode ray tube and this results in variations in brightness of the point of light appearing on the screen, which correspond to variations in the strength of the signal. The position of these points of light is made to represent geometrically their point of origin within the body so that a composite two dimensional picture can be obtained at any desired level.

b. Ultrasonic Placentography

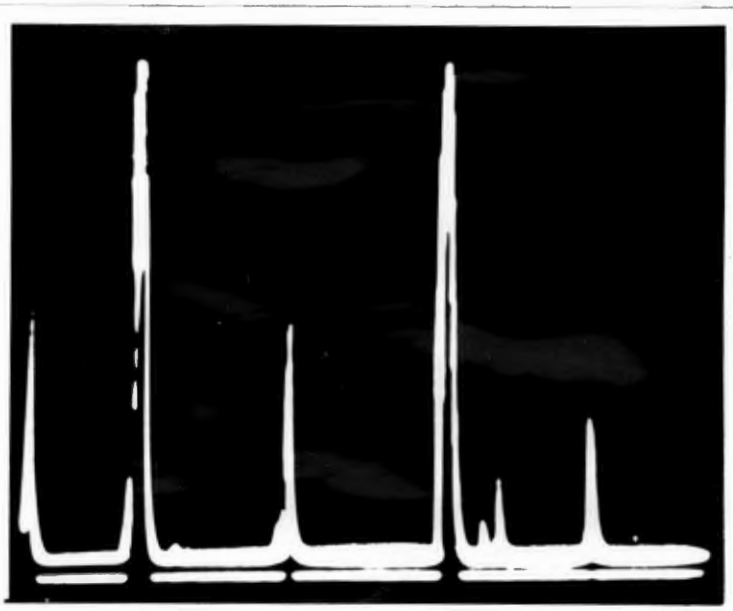
This technique of placental localisation is now standard in our unit.

Figure 11



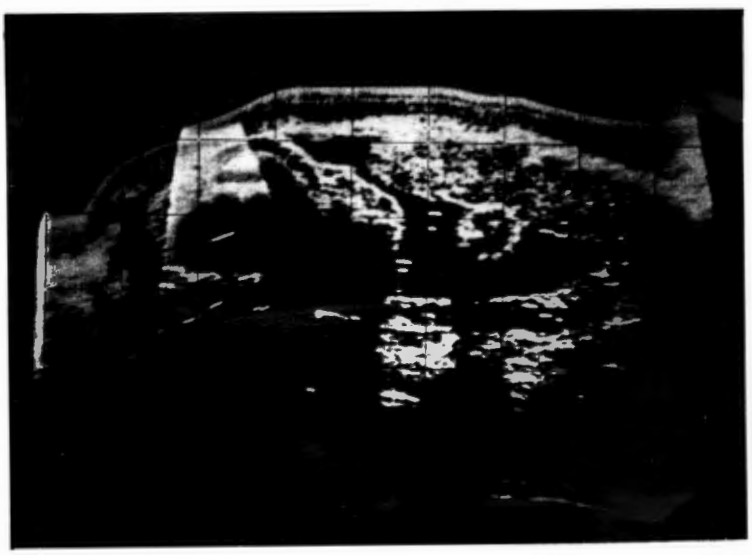
A picture obtained of the fetal skull in two-dimensional display (B-scan) as obtained on the Diasonograph.

Figure 12



A one-dimensional display (A-scan) of the fetal biparietal diameter

Figure 13



A two-dimensional ultrasonic display indicating the presence of the placenta. The firm line indicates the interface between placental amnion and liquor amnii.

The method used was that of Donald and Abdulla (1968) based on the work of Gottesfeld, Thompson, Holmes and Taylor (1966).

The patients were all requested not to empty their bladders prior to ultrasonography as the transonic bladder provides a very valuable landmark, even in the last trimester (Donald, 1968). With the patient in the supine position, her abdomen liberally covered with olive oil, and at low gain amplification of the ultrasound, the uterus was scanned longitudinally from left to right using the B-scan display. The uterus was then scanned transversely from the fundus to the symphysis.

At low gain the placenta shows as a clear area, which begins to fill with "speckles" as higher gain is used. In most instances the fetal surface of the placenta can be seen as a definite firm white line. In this manner both the position and extent to the placenta can be determined with an accuracy of 94% (Campbell and Kohorn, 1968) or higher (Gottesfeld et al, 1966).

Any patient in whom an anteriorly situated placenta was diagnosed by this method was excluded from the study group on the grounds that the risk of amniocentesis was too great.

Every patient in the study group had ultrasonic placental localisation done before the first amniocentesis was performed. A very careful note of the placental site was made in each patients' notes and referred to prior to each subsequent amniocentesis.

c. Ultrasonic Cephalometry

Following the work of Donald and Brown (1961) and more recently Campbell (1969), the antenatal measurement of the fetal

biparietal diameter by ultrasound has come to be accepted as a means of predicting fetal maturity. In an earlier report, Campbell (1968) reported that from the 20th to the 30th week of pregnancy the biparietal diameter values were confined within narrow limits for any given week of gestation, and that measurement during this period could be valuable in the prediction of fetal maturity.

Although it is accepted that the Standard Deviations of Mean measurements for given weeks of gestation after the 30th week of gestation become larger, and that the increase in the biparietal diameter does not bear a linear relationship to time after this period, it was felt that this parameter might provide at least part of the answer to the problem of gestational ageing.

By using both the A and B scan display systems as described above, biparietal diameter measurements were made on each patient on three occasions. As with amniocentesis, an attempt was made to obtain these measurements at 32, 35, and 37 weeks of gestation.

The correct measurement of the diameter by means of electronic calipers on the A-scan is assured by initial orientation of the head in two dimensional B-scan display. This enables accurate positioning of the probe.

Prior to the start of this study, the electronic caliper was accurately calibrated by an agent for Nuclear Enterprises Ltd. Donald (1968) maintains that with experience, accuracies to within 0.5mm are obtainable.

All the measurements in this series were made by a qualified gynaecologist who received his ultrasonic experience in Professor Donald's unit in Glasgow.

All readings were recorded in the patient's research folder and not in her obstetric notes.

13. PAEDIATRIC ASSESSMENT OF THE NEWBORN

Until fairly recently the postnatal assessment of fetal maturity rested upon birth weight and length (Lubchenco, Hausman, Dresler and Boyd, 1963). Paediatricians have now become aware of the unreliability of these parameters and have sought alternative methods of assessing gestational age. The clinical method of assessing the gestation age of newborn infants in the Cape Town unit is that of Dubowitz, Dubowitz and Goldberg (1970). This method and others (Farr, Mitchell, Neligan and Parkin, 1966) were developed out of the need to differentiate the short gestation from the light-for-dates infants.

All European infants born of 'research' mothers were assessed by the Senior Paediatrician at the hospital concerned within forty-eight hours of delivery and all non-European infants born of 'research' mothers similarly assessed at the other hospital concerned. Both paediatricians were Senior Consultants and used exactly the same method.

The method used makes use of both a neurological assessment and a critical evaluation of certain external physical characteristics (Appendix D). Using both these methods and adding the scores to give a total score, Dubowitz et al (1970) found a greater accuracy than when either of the neurological or external characteristics were used alone. Their accuracy was claimed to have 95% confidence limits of 2.04 weeks. The paediatricians involved in this study had previously shown an accuracy of 2.16 weeks with 95% confidence limits (Harrison, 1970).

Paediatric assessment of gestational age was held to be

essential to the integrity of this study as it was the only safe, reliable and practical method of checking the validity of the dates of the L.N.M.P's as provided by the patients. Any patient in whom the discrepancy, between the 'paediatric gestational age' and that calculated from the L.N.M.P., exceeded two weeks was excluded from the final number of patients whose data was to be analysed.

14. NEONATAL WEIGHT FOR AGE

Although a concerted effort had been made to select normal patients for inclusion in the survey, it was essential that each infant be normal before the patient's data could be accepted as that obtained from a normal uncomplicated pregnancy.

In the method of assessment used by the paediatricians (see section 13. above) an obvious departure from normal would have been discovered. It was felt, however, that each infant would have to conform to a rigid weight-for-age standard, to exclude all possible cases of fetal malnutrition. The figures used for comparative purposes were those of Gruenwald (1969) and were matched for sex and parity. Any infant in this series falling below one standard deviation from the mean was regarded, for the purposes of the analysis of data, to be 'light-for-dates'.

Diagnosis of fetal malnutrition based upon growth retardation alone (low birth weight for gestational age) is open to at least three criticisms:

i. although low birth weight for gestation may represent growth retardation due to intra-uterine malnutrition, it may also be intrinsic in origin, representing the lowest part of the normal distribution curve for fetal growth potential.

ii. a second criticism is that this criterion depends upon accurate knowledge of gestational age and, therefore, upon the menstrual history.

iii. a third handicap of this method of diagnosis is the lack of accepted standards for birth weight at each week of gestational age, based on live births of the same race and nationality living at the same altitude as those to be investigated for fetal malnutrition.

Nevertheless, it was felt that a rigid standard of normality had to be applied in order that the data obtained could be regarded as valid and representative of normal.

CHAPTER 4

STATISTICAL ANALYSES

All statistical analyses on the data obtained in this study were performed by the Division of Medical Statistics and Epidemiology of the South African Medical Research Council in Pretoria.

The complex problem of analysing all the results obtained was referred to the Department of Mathematical Statistics at the University of Cape Town as well as to Mr. W.Z. Billewicz of the British Medical Research Council Reproduction and Growth Research Unit, Newcastle-upon-Tyne, England. The conclusion arrived at by these experts in the field of statistics was that the technique of stepwise discriminant analysis was the most satisfactory method of handling the data. Professor Troskie of the University of Cape Town referred the candidate to the Director of the abovementioned unit in Pretoria as the University of Cape Town Computer Centre was unable to cope with the type of analysis required.

1. TIME GROUPS

A total of 23 variables were measured and in addition to these, two additional variables were calculated, viz. the difference between maternal and liquor urea and the difference between maternal and liquor true creatinine. Twenty-one of the 25 variables were measured at three stages of each pregnancy and, as has already been mentioned, these stages were approximately 32, 35 and 37 weeks. The last ten weeks of pregnancy were divided into three intervals because prediction of gestational age, accurate to within one week, was felt to be clinically inappropriate and impossible within the scope of this study. The three intervals selected were 30 - 33 weeks,

34 - 36 weeks and 37+ weeks.

TABLE I

The Mean, Standard Deviation (S.D.), Range, Maximum and Minimum of the duration of pregnancy in weeks when amniocenteses were performed

TIME GROUPS (weeks)	TIME IN WEEKS				
	MEAN	S.D.	RANGE	MAXIMUM	MINIMUM
30 - 33	31.7	0.5	2	33	31
34 - 36	34.9	0.6	2	36	34
37+	37.1	0.4	2	39	37

Analysis of the data was performed on the time basis of completed weeks of gestation although the duration of pregnancy in days was known. Using 'completed weeks', the majority of the information fell into one of the above three time intervals.

Where more than three measurements of one variable were made e.g. uterine volume, only one of those measurements falling into each of the time intervals was used for analysis. Where more than one measurement fell into a particular time interval, the measurement used for computation was that obtained in the same completed week as the rest of the variables. On the few occasions when this was not available viz. the patient had her amniocentesis in the 32nd week of pregnancy but was only examined clinically in the 31st and 34th weeks, the measurement obtained in the nearest week within that time interval was used.

The level of statistical significance used in the following analyses was $P = < 0.05$.

2. THE FRIEDMAN TWO-WAY ANALYSIS OF VARIANCE BY RANKS

The Friedman two-way analysis of variance was applied to each variable independently (Siegel, 1956). The Friedman two-way analysis of variance is applicable when data in the form of k matched observations are at least ordinal, and tests the null hypothesis that the k observations had been drawn from the same population. In this case k = 3, viz. the three times at which observations were taken on a patient. Where the number of patients included was N, the data for a certain variable is cast in a two-way table as shown, i.e. N rows and k columns.

TABLE II

The method of application of the Friedman Two-way Analysis of Variance by Ranks to an individual variable

PATIENTS	TIME		
	30 - 33 weeks	34 - 36 weeks	37+ weeks
PATIENT 1	$X_{1,1}$	$X_{1,2}$	$X_{1,3}$
2	$X_{2,1}$	$X_{2,2}$	$X_{2,3}$
.	.	.	.
.	.	.	.
.	.	.	.
.	.	.	.
.	.	.	.
N	X_{N1}	X_{N2}	X_{N3}

X_{ij} represents the value of the variable for the i^{th} patient taken at the j^{th} time epoch. In order to perform the Friedman Test on these data, each row is ranked; that is the

smallest value is replaced by 1, the next largest by 2 and the largest by 3. Each column is then added to give R_k, the ranked total. If the patients' scores were independent of the three times at which the observations were taken the totals would vary from one column to another. The Friedman Test determines whether the rank totals differ significantly. For this test the following statistic is computed:-

$$\chi r^2 = \frac{12}{Nk\ (k+1)} \sum_{j=1}^{k} (R_j)^2 - 3N\ (k+1) \qquad (1)$$

when N = number of patients

k = number of observations on each patient

R_j = sum of ranks in j^{th} column

If the value of χr^2 is greater than a certain prescribed critical value the null hypothesis that the observations are independent of time is rejected at a certain level of significance.

3. STEPWISE DISCRIMINANT ANALYSIS

A large number of parameters were measured on three occasions or more on 47 pregnant women for whom the age of the fetus was known. The problem was to pick out those parameters which could best be used to estimate the age of the fetus for future cases where this was not known.

Since more than one measurement was obtained from each patient at different times, the sample observations could not be considered to be independent of each other. The stepwise discriminant analysis has a basic assumption that the measurements are independent and, therefore, the results obtained by this method of

statistical analysis must be viewed with caution. An attempt was made to ascertain what sort of result might have been obtained had dependence between time periods not been present. The cases were divided randomly into three groups and within a group the observations at only one time interval used.

a. Discrimination

The problem in discrimination is as follows:-
Given knowledge that g groups of patients exist (three age groups in this case) and given a set of observations on a sample of people from these groups, a rule is devised which will enable correct assignment of some new person to the group to which she belongs.

In the discriminant analysis approach we seek for each group a discriminant function of the form:

$$X_i = C_{io} + C_{i1}x_1 + C_{i2}x_2 \ldots\ldots\ldots + C_{iN}x_N \quad (2)$$

where in C_{ij} : $i = 1.....g$, $j = 0$, and $1.....N$ are constants

and in x_i : $i = 1.....N$ is the i^{th} variable measured

The constants C_{io} —— C_{iN} are determined so that X_1 - - - X_g will differ maximally for each group, i.e. the ratio of variance within groups to total variance will be a minimum.

In order to make use of these g functions to classify a new case, the values obtained for N variables measured must be substituted in each of the g equations (2) in turn and the X_i value which is the greatest, e.g. X_k, selected. The new case would then be placed in the k^{th} group.

When it is desired to know the probability of the new case falling into any one of the groups, for example group m, this probability is denoted by Pm.

$$\text{Then } Pm = \frac{\exp X_m}{\sum_{i=1}^{g} \exp X_i} \qquad (3)$$

Note $P_1 + P_2 \ldots\ldots\ldots\ldots + P_g = 1$ and P_k will be the maximum of $P_1 \ldots\ldots P_g$ if X_k is the maximum of $X_1 \ldots\ldots X_g$.

b. Stepwise Discrimination

As some of the N variables measured on a subject may have contributed little or nothing to the discrimination, it was necessary to select that subset which would best assist the author in allocating a patient to her correct group. Firstly, the variable which, when used alone, provided the best discrimination between the groups was selected, i.e. which minimised the ratio of variance within groups to total variance. In the next step the variable which, when used in conjunction with the first selected variable, gave the best discrimination between the groups was added. This process was continued in successive steps and at each step a variable which had already been selected may have been excluded, if, due to the inclusion of other variables, its contribution had now become unimportant. Variables were no longer added to the function at the step where the inclusion of any of the variables not already included would not have significantly improved the discriminant function. As previously mentioned, the level of $P = <0.05$ was used throughout this analysis.

In using the discriminant technique it is necessary to

have at least three more cases than the number of variables. It is, however, preferable to have five or more times as many cases as the number of variables, as the more cases there are, the more valid the results. This implies that in this analysis, the number of cases, even where a subsample was used, was sufficient.

For the purposes of the Friedman two-way analysis of variance and the stepwise discriminant analysis, an I.B.M. 360 Model 65 computer was used.

4. COEFFICIENTS OF CORRELATION AND REGRESSION LINES

A model 9100B Hewlett-Packard Calculator was used by the author to calculate the coefficients of correlation and plot the regression lines for two-way analysis of variables. The coefficient of correlation was calculated by employing the equation:

$$r = \frac{\sum (x - \bar{x})(y - \bar{y})}{\sqrt{(x - \bar{x})^2 \sum (y - \bar{y})^2}} \qquad (4)$$

The regression lines were drawn by making use of the method of least squares, i.e. the line was fitted to minimize the sum of the squares of the vertical distances between the plot points and the line.

5. MEAN AND STANDARD DEVIATION

The mean ($\bar{X}$) was obtained by dividing the sum of all the values ($\sum X_i$) by the number of values (n) i.e.

$$\bar{X} = \frac{\sum X_i}{n} \qquad (5)$$

The standard deviation (S.D.) as a measure of the

dispersion of values around the mean, was calculated by the following equation:

$$S.D. = \sqrt{\frac{\sum(X_i - \bar{X})^2}{n - 1}} \qquad (6)$$

6. STANDARD ERROR OF THE MEAN

This statistic was used to test the significance of differences of means of the results obtained from normal and 'light-for-dates' pregnancies.

This statistic was obtained from the equation:

$$\frac{S.D.}{\sqrt{n}} \qquad (7)$$

CHAPTER 5

RESULTS

INTRODUCTION

To avoid repetition, the customary chapter covering the historical aspects pertaining to this research work has not been included in this thesis. Instead an historical introduction to each section has been included before the results are reported.

The author feels that a summation of the results obtained and a review of the relevant literature is preferable to two dissociated yet intimately linked aspects.

In spite of the statistical analyses being performed upon results obtained expressed in completed weeks of gestation, all the figures in the following section have been plotted with the results obtained at the specific point in time at which the samples etc. were taken, i.e. a result obtained at 37.5 weeks was plotted as such, yet was regarded as a result obtained after 37 completed weeks for the purposes of analysis. This differentiation was made to simplify the plotting of the graphs, as it would prevent all the same figures being plotted one on top of the other.

All the results obtained in this study are tabulated in Appendix E.

1. MATERNAL NORMALITY

In a previous study of 4,552 primigravid singleton hospital live deliveries in the Cape Town Unit in 1970 (Bennett, 1972a) it was found, on the basis of comparisons of means and standard deviations, that there was a significant difference between the European and non-European patients. In general terms the non-European primigravidae were younger, shorter in stature, lighter in weight at their first visit and produced smaller babies than did their European counterparts. That they were a different race certainly does not prove an aetiological relationship, in fact socio-economic factors almost certainly play a more dominant role than does ethnic group.

It was in order to eliminate, as far as was possible, the role played by socio-economic factors, that the patients were selected according to the criteria already discussed (Chapter 2, section 1, page 4). In general terms, pregnant women in Cape Town who can afford the services of a private practitioner, do so. This leads the vast majority of non-Europeans to seek hospital delivery, whereas the Europeans delivered in hospital either cannot afford a private practitioner or have some pathological state complicating their pregnancies which require intensive hospital supervision e.g. diabetes mellitus, cardiac disease etc. It is thus expected that those patients at least risk from socio-economic influences are not available for study by full-time hospital staff. The 'average' European patient delivered in the Cape Town unit is, therefore, of a lower socio-economic standing than the average European delivering in Cape Town generally. The ratio of private to hospital deliveries in Cape Town is estimated to be at least 10:1. As the vast majority of non-Europeans deliver their babies in hospital, it is to be expected

that the socio-economic standing of the 'average' hospital delivered non-European approximates that of the average for the general non-European population.

TABLE III

The mean, age, height, booking weight and birth weight of infants for the two races in the two groups of patients

Group of Patients	Number of Patients	Age (yrs)	Height (cm)	Booking Weight (kg)	Birth Weight of infant (gm)
U. E.	750	21.3	162.7	63.2	3065
U. N-E.	3802	20.6	155.5	56.8	2918
R. E.	24	21.5	161.5	58.7	3069
R. N-E.	23	22.0	159.5	57.4	2987

E : European

N-E : Non-European

U : Primigravid singleton live deliveries in the Cape Town unit in 1970

R : Patients in the research group

From Table III it would appear that the attempt that was made to reduce the influence of socio-economic factors to approximately the same level for the two race groups, was successful. The figures would seem to indicate that the criteria by which the patients included in this study were selected, have in some measure reduced the gap between the race groups. Naylor and Myrianthopoulos (1967) found a direct association between birth weight and socio-economic status and

Gruenwald (1966) concluded that the differences between racial groups were in large measure socio-economic rather than racial. The above figures tend to support these authors. The fact that the mean birth weight of the European patients in both these studies is below that found for Cape Town by Malan et al (1967), is probably an index of the increasing number of patients being delivered by private practitioners, leaving the lower socio-economic status patients to deliver in hospital.

Based solely upon the routine assessment of patients as performed at the antenatal clinics in our unit, no patient in this research group manifested any signs of deviation from accepted clinical norms of pregnancy. As will be seen later, there were some patients who showed an obvious deviation from the mean for certain of the parameters being measured, but as these were research methods, they were only assessed and analysed when the last patient had delivered. The analysis of the measurements being made in this research project was delayed until all the patients had delivered because (i) the author did not know what the norm was until analysis had been undertaken (ii) the author wanted to avoid possible influences upon future measurements, and (iii) it was felt that all the data had to be analysed before he could draw any conclusions.

It has, therefore, been shown that socio-economic influences have been reduced in the non-European patients studied, to very nearly the level of the European patients. The pregnancies which yielded the data for final analysis were clinically normal and the infants resulting from these pregnancies were normal within specific limits as discussed earlier.

As has already been mentioned, all these mothers and their babies returned for a postnatal check-up some 6 weeks after delivery. At this visit each patient was again fully examined by the author and

each baby examined by one of the two paediatricians concerned. All the patients were found to be generally and gynaecologically well and no baby manifested any abnormal features.

2. NORMALITY OF INFANTS

At the outset it was decided that a normal healthy patient should produce a normal healthy baby after an uncomplicated pregnancy before any of the measurements obtained during that pregnancy could be accepted for analysis. The criteria for selection of a normal healthy patient have already been enumerated (Chapter 2, section 1, page 4) and will not be discussed further. An uncomplicated pregnancy was defined as one in which no clinical evidence of a pathological state, e.g. hypertension, albuminuria, antepartum haemorrhage, clinical suspicion of intra-uterine growth retardation etc., was found during that pregnancy.

A. Method of Delivery

It was initially considered necessary for a normal pregnancy to end in a spontaneous vaginal delivery. It was soon realised, however, that a Caesarean section performed, for example, on a primiparous patient, after the onset of spontaneous labour for a breech presentation, could not in any way be considered as evidence of a pathological state. The method of delivery was only accepted as indicative of a pathological state if an operative or assisted delivery was performed for fetal distress, except when cord compression was the cause of the distress. Towards the end of the series the author acquired the technique of caudal block anaesthesia and the last 10 patients received this form of analgesia during labour. This accounts for the relatively high incidence of forceps deliveries.

The methods of delivery and their indications are shown in Table IV.

TABLE IV

The method of delivery of the 47 patients studied and the indications for operative intervention

Method of Delivery	Indication	No. of Cases
Caesarean Section	Abruptio Placenta*	1
Caesarean Section	Prolapsed cord	1
Forceps	Persistent Occipito-posterior position	1
Forceps	Caudal block	8
Vacuum Extraction	Cord compression*	1
Spontaneous Vaginal Delivery	-	35

* These two cases are fully discussed elsewhere (page 144).

Following delivery, each infant was weighed and measured and then assessed by the paediatrician concerned for both congenital abnormalities and its gestational age. Apgar scores were not regarded as an index of the normality of the pregnancy for the same reasons as discussed under the method of delivery.

Provided that the infant had no congenital malformations, showed normal responses, had a birth weight of not less than one standard deviation below the mean for its gestational period, and had a paediatric assessment of gestational age within $\pm$ 8 days of its calculated age, it was accepted as normal for the purposes of this study.

B. Assessment of Weight for Age

For the assessment of weight for age, curves were drawn

using the figures of the British Perinatal Mortality Survey (Butler and Alberman, 1969) for primiparae. These figures were used because they cover a whole population and were considered the most complete available. Two sets of curves were used because of the known fact that the birth weight of girls tends to be lower than that of boys (Lubchenco et al, 1963), a fact confirmed by the curves derived after 36 weeks.

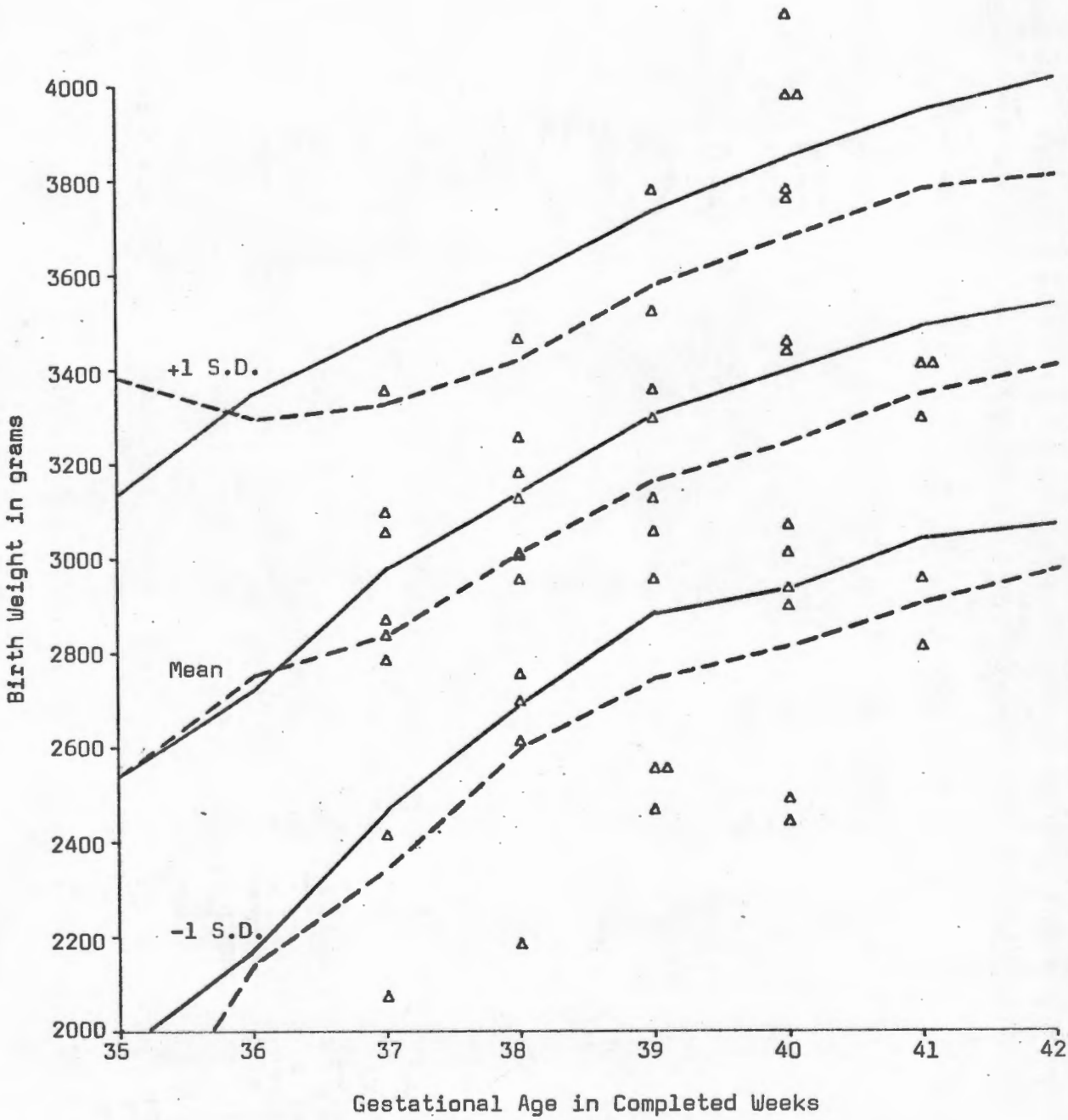
As can be seen from Figure 14 there were eight infants who at birth, weighed less than one standard deviation below the mean for age. The information obtained in this study from these eight pregnancies was separated from the rest of the information and analysed separately, since by a previous definition they were 'light-for-dates' infants. Although, by the same definition, those pregnancies resulting in infants more than one standard deviation above the mean should have also been excluded, it was decided to retain them in the group for analysis. The reason for this was that on clinical grounds the problem of wrong dates versus intra-uterine growth retardation always arises when a fetus is though to be smaller than its expected size for the duration of pregnancy, as calculated from the L.N.M.P.

Figure 15 shows the birth weights plotted against the crown-heel length in centimetres, and the ratio of weight to length against age at delivery in Figure 16.

C. Paediatric Assessment of Gestational Age

Dubowitz et al (1970) claim an accuracy of 2.04 weeks within 95% confidence limits using their combined scoring system. Farr et al (1966), using only external physical characteristics, claimed an accuracy of 2.4 weeks within the same confidence limits. As will be seen from Figure 17 the 95% confidence limits of

Figure 14

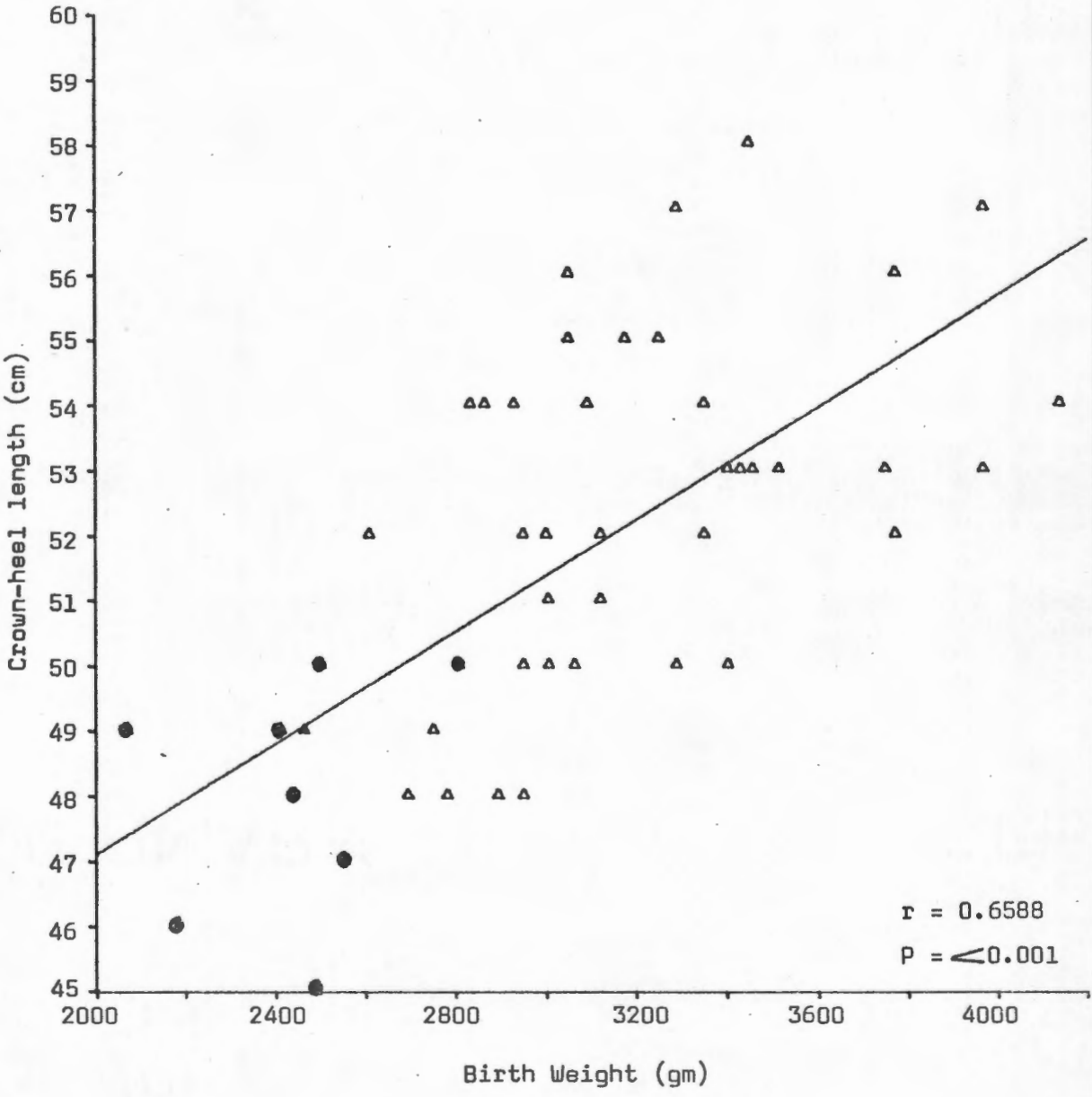


Birth weight plotted against gestational age at delivery in completed weeks calculated from the L.N.M.P. The means ±1 standard deviation were those found for primigravidae in the British Perinatal Mortality Survey (Butler and Alberman 1969)

----- female

——— male

Figure 15

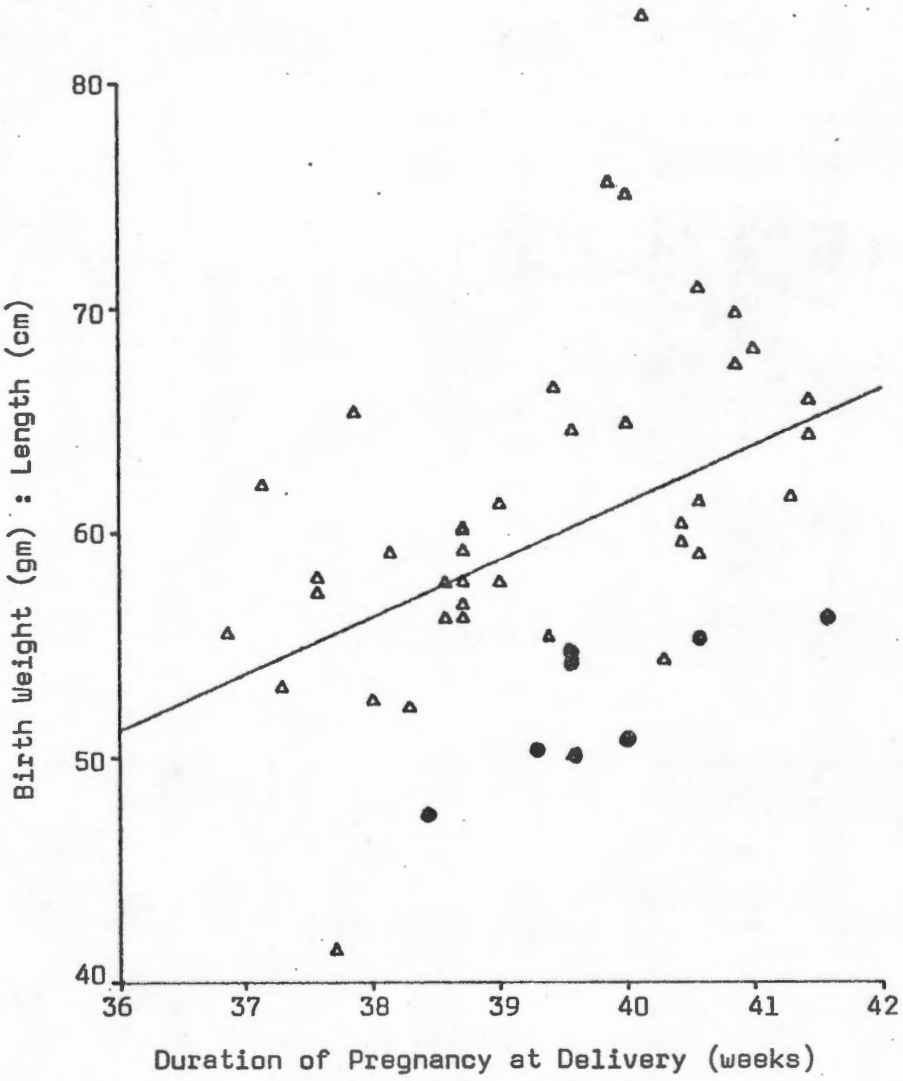


The relationship between crown-heel length and birth weight

△ Normal infants

● 'Light-for-dates' infants

Figure 16

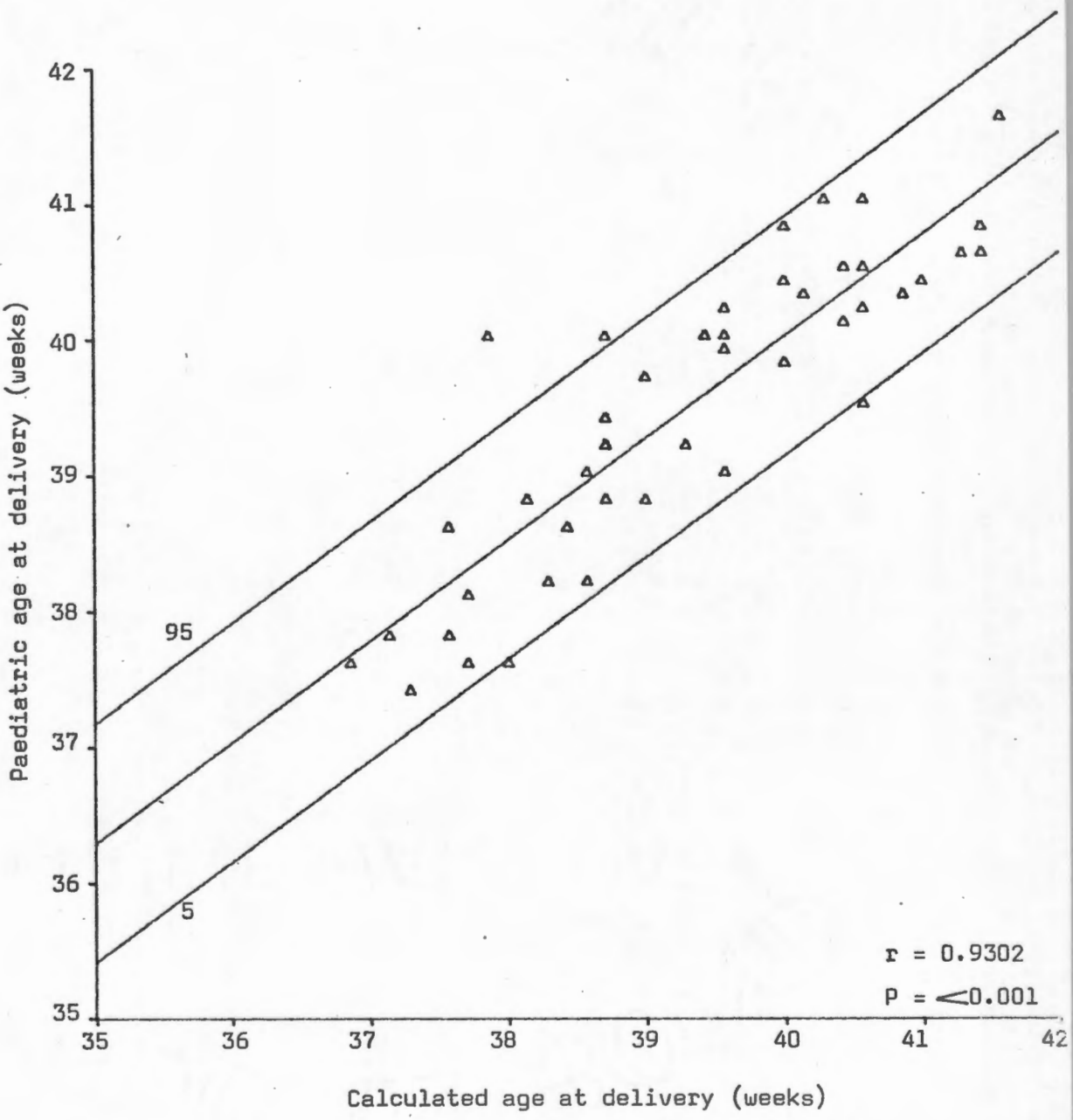


The ratio of birth weight to length by week of gestation

△ Normal infants

● 'Light-for-dates' infants

Figure 17



Paediatric assessment of gestational age plotted against the gestational age calculated from the L.N.M.P.

2.16 weeks was nearly mid-way between the above two figures. This may well be due to the fact that instead of one paediatrician assessing every infant it was necessary to use two because of the different hospitals in which the two racial groups were delivered. The correlation coefficient for the paediatric assessment against gestational age was the same as found by Dubowitz et al, i.e. 0.93. It was unfortunately not possible within the framework of this study, to match the two paediatricians against each other on the assessment of the same infants. In a previous study involving the same two paediatricians the maximum difference in total scores using the same infants was three points, (Malan, 1971). In view of this previous finding and the close correlation between the results obtained in this study and those of Dubowitz et al (1970), the author feels justified in accepting the paediatric assessment of gestational age as confirmatory evidence of the accuracy of a patients 'dates'. No patient in this research series was found, on the above criterion, to have been incorrect when giving the date of her L.N.M.P.

In Figure 17 the known gestational age and the paediatric assessment of gestational age are plotted against each other.

D. Puerperal Examination

Each infant returned for a full examination by the same paediatrician as initially examined it six weeks after delivery. A full general and neurological examination was performed. None of the babies showed any deviation from normal in respect of growth and development subsequent to delivery.

3. PARAMETERS EXCLUDED FROM ANALYSIS

Certain of the parameters measured were not subjected to statistical analysis as it was felt that either they were not actually parameters of gestational age or would detract from the validity of the final results.

A. Maternal Weight

This was excluded simply because it can have no bearing whatsoever upon the duration of pregnancy. Obviously women vary in their pre-pregnant weights by more than 45 kilograms and as such, no single reading or even succession of readings can provide any information regarding the duration of pregnancy. That a pregnant patient will put on weight during the course of her pregnancy, all else being equal, is appreciated by all who deal with such patients. Factors such as the rate of increase in weight gain during any single pregnancy are clinically useful guides to the state of wellbeing of the fetus but not to the gestational age of that fetus.

B. Clinical Height of Uterine Fundus

As was described earlier (Chapter 3, section 1, page 11), a clinical evaluation of the fundal height was made at every antenatal visit of each patient. The initial object was to have one senior consultant assess each patient without prior knowledge of the calculated duration of pregnancy, in an attempt to exclude observer bias. In the early part of this project this unbiased assessment was possible but as the number of patients grew it became impracticable and was abandoned. Each patient was, thereafter, assessed solely by the author and although a conscientious

attempt was made to exclude any such bias as might have occurred due to the author knowing each patient as well as he did, observer bias could not honestly be excluded from having played a role in the assessment of the fundal height. It was felt that this parameter should, therefore, be omitted from the statistical analysis.

It has been well shown by Beazley and Underhill (1970) that the fundal height as a parameter of gestational age is liable to large errors, even in experienced hands, when a single assessment is made with no information about the patient available.

C. Radiological Assessment of Gestational Age

Radiological assessment of gestational age was excluded from the analysis because it was not a serially estimated parameter. All the other parameters were estimated upon three or more occasions and could thus be analysed against time. Radiological assessment could, therefore, only be classified as correct or incorrect. Those cases in which the radiologist was incorrect as judged by the known duration of pregnancy at the time of X-ray, were subdivided further into those in which the radiologist reported an age in advance of the known age and those in which he reported an age less than the known age.

Radiological reports were, therefore, classified as:

i. under estimated

ii. correctly estimated

iii. over estimated

and will be discussed as a separate section.

4. CLINICAL PARAMETERS

A. Gestational Age calculated from the first day of the patients' last normal menstrual period

The actual duration of pregnancy is obviously from the time of conception to the time of delivery. In a study of 1336 women, having spontaneous deliveries whose last ovulation had been recorded by a rise in basal body temperature, Guerrero and Florez (1969) found the time between the thermal shift and delivery to be 265 (S.D. ± 11.9) days. As the time between menstruation and ovulation is variable, gestational age is in practice dated from the start of the last normal menstrual period (L.N.M.P.).

Lind (1970) concludes that the most valuable single piece of information the clinician can have by which to assess the functional development of the fetus is an accurate date for the onset of the last menstrual period from a woman with a normal regular cycle. He admits, however, that such information may not be available, or it may be misleading. The increasing use of oral contraceptives can lead to withdrawal bleeding being interpreted as true menstruation and, therefore, confusing the clinician ab initio.

As has already been mentioned, little reliance can be placed on the date of the L.N.M.P. given by patients delivering in our unit in Cape Town and the author, whilst accepting Lind's conclusion, has reservations about its application in Cape Town.

The patients in the research group, all had regular cycles, were certain of the date of their L.N.M.P. and were generally reliable historians. Nevertheless, the author awaited the paediatric assessment of the gestational age of the baby before

accepting the calculated gestational age as the basis for analysis of the parameters measured.

Using this form of cross-check, none of the patients in the study group were found to have made an error when giving the date of their L.N.M.P.

B. Estimation of Uterine Size at the First Visit

It has long been known (Lind, 1970) that the earlier in pregnancy that the uterine size is assessed, the more accurate is the assessment of the duration of pregnancy.

In the research group of patients, the mean gestational age at the time of booking was 16.5 (S.D. ± 3.7) weeks and the assessment of uterine size at that visit was 16.6 (S.D. ± 3.9) weeks.

TABLE V

Clinical Assessment of Gestational Age at First Visit

	Mean	S.D.
Calculated gestational age	16.5	3.7
Clinical assessment of uterine size	16.6	3.9

C. Date of Quickening

Kraus and Hendricks (1964) concluded that the tremendous range in time at which the pregnant patient subjectively feels fetal movements for the first time, makes this parameter a useless one as an indicator of the duration of pregnancy. They found that they were unable to correlate the date of quickening with the time interval to delivery.

In the 47 patients studied by the author, the mean duration of pregnancy at the time of quickening was 134 (S.D. ± 14.2) days and compares well with that found by Kraus and Hendricks in a larger series of primigravidae.

TABLE VI

Mean and Standard Deviation of the date of Quickening

	Mean	S.D.
Kraus and Hendricks (1964)	134.9	± 13.2
Gestational age research patients	134.0	± 14.2

D. Clinical Assessment of the Height of the Uterine Fundus (H.O.F. clin.)

Every standard textbook of obstetrics has a diagram in which the H.O.F. clin. is related to abdominal landmarks in terms of weeks of gestation. These landmarks are anatomical ones and the usual three are the pubis, the umbilicus and the xiphisternum. The level of the uterine fundus is often measured in finger breadths above or below these anatomical points.

Objections to this method of assessing the duration of pregnancy are:

i. the distances between any two of these three anatomical points are not constant.

ii. many factors influence the size of the conceptus at any given period of gestation apart from the gestational age.

iii. obviously the size of the uterus is related to the size of the conceptus it contains and this in turn

is related to the length of gestation, but these correlations are not close enough to make anything but the crudest approximation to gestational age.

Beazley and Underhill (1970) found that the range of measurements of the length between symphysis pubis and xiphisternum in 233 women was 25 - 43cm,and the position of the umbilicus varied from 11.5 - 19cm above the symphysis pubis.

Using the traditional landmarks, the author correlated the uterine size with gestational age and found a good correlation in those patients who produced normal infants. The correlation was poorer when the 'light-for-dates' pregnancies were considered (Appendix E).

As has been explained, observer bias could not be excluded and thus this parameter was not analysed further.

E. Height of the Uterine Fundus in centimetres above the Symphysis Pubis

Table VII shows the mean and standard deviation in centimetres of the height of the fundus above the upper border of the pubic symphysis for both normal and 'light-for-dates' pregnancies. From Figure 18 a fairly wide scatter can be observed in the level of the fundal height for any given week of pregnancy. A much wider range is apparent, however, when a single measurement is taken and gestational age is then sought. At a level of 34cm the range is from 33 - 40 weeks in the normal pregnancies. When considering the 'light-for-dates' pregnancies, the maximum range of 29 - 38 weeks is found to correspond to a fundal height of 29cm.

F. Uterine Volume

In a recent study from Cape Town (Baillie, 1972), the imprecise clinical measurement of uterine volume was found to

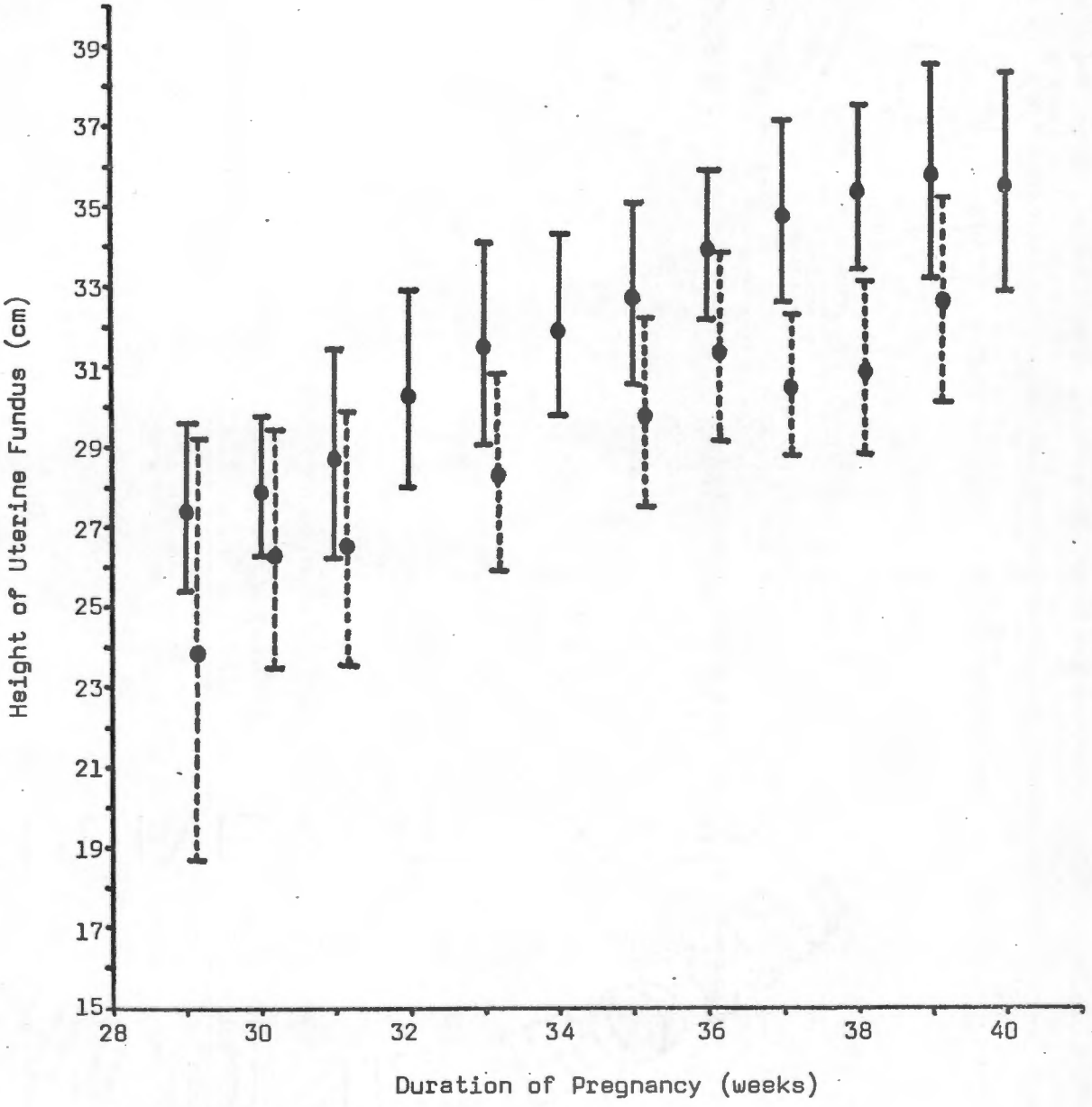
TABLE VII

MEAN AND STANDARD DEVIATION OF THE HEIGHT OF THE UTERINE FUNDUS IN CENTIMETRES ABOVE THE SYMPHYSIS PUBIS FOR EACH COMPLETED WEEK OF GESTATION

(Only one reading was obtained from 'light-for-dates' pregnancies at 32 and 34 weeks and none at the 40th week).

Gestation in weeks	Normal Infants			'Light-for-dates' Infants		
	Number of cases	Mean Fundal Height (cm)	Standard Deviation	Number of cases	Mean Fundal Height (cm)	Standard Deviation
29	13	27.4	2.1	2	23.8	5.3
30	18	27.9	1.7	5	26.3	3.1
31	20	28.7	2.6	7	26.5	3.2
32	19	30.3	2.4	1	-	-
33	21	31.5	2.4	6	28.2	2.5
34	25	31.9	2.2	1	-	-
35	17	32.7	2.2	7	29.7	2.3
36	33	33.9	1.8	6	31.3	2.4
37	29	34.7	2.2	6	30.4	1.7
38	26	35.3	2.0	5	30.8	2.1
39	16	35.7	2.6	4	32.5	2.5
40	8	35.4	2.7	-	-	-

Figure 18



Mean ± 1 standard deviation of the height of the uterine fundus above the symphysis pubis for each week of pregnancy after 29 wks.

Normal infants

'Light-for-dates' infants

correlate well with the actual state of the baby at delivery. This was especially true when the percentage volume increase was considered. There was, however, a correlation between uterine volume and gestational age and the purpose of including this parameter in this study was to evaluate this correlation.

Table VIII shows the mean and standard deviations of the uterine volume in cubic centimetres for each completed week of gestation studied in both normal and 'light-for-dates' pregnancies. In Figure 19 these values have been plotted against the duration of pregnancy and a wide range for any given week of pregnancy is observed. A very large range for the gestational age for any particular uterine volume measurement is apparent. The explanation for this may lie in the unknown variation in abdominal wall thickness between patients i.e. measurements a and b in Figure 1.

A correlation coefficient of 0.6858 is obtained when birth weight is plotted against the last measurement made of the uterine volume, prior to delivery. As only one infant had an Apgar score of less then seven at delivery, no correlation between uterine volume and the state of the baby at delivery, could be made.

A volume of more than 5,500cm^3 was found to indicate a birth weight of over 2,600 grams although the converse is not true (Figure 20).

G. <u>The State of the Cervix</u>

Grading of the state of the cervix in the last 10 weeks of pregnancy necessitated repeated vaginal examinations. These were not done as sterile procedures but a sterile glove was always worn and an antiseptic cream used as a lubricant. The procedure was carefully explained to each patient and the author was as gentle as

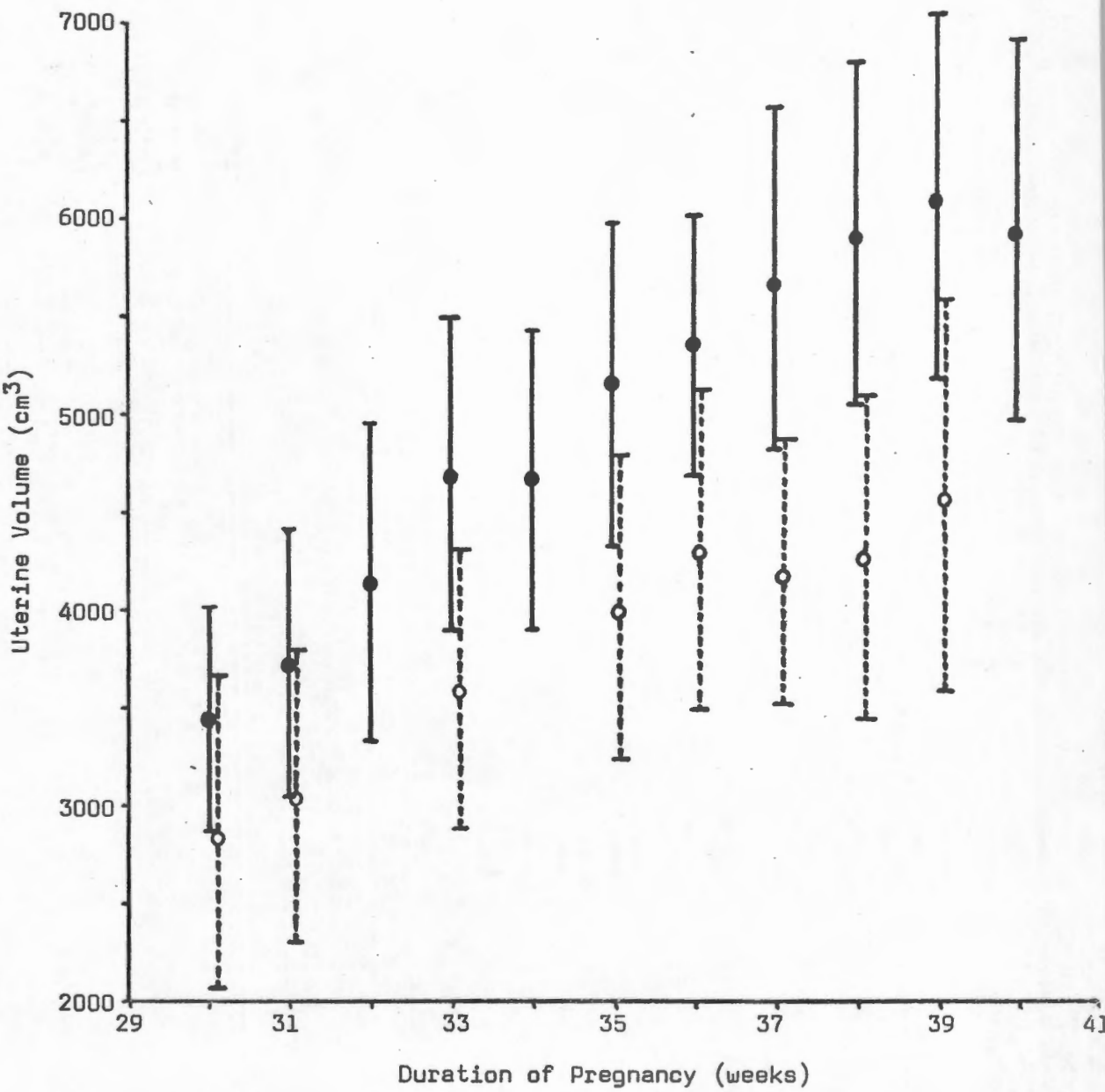
TABLE VIII

MEAN UTERINE VOLUME AND STANDARD DEVIATIONS IN CUBIC CENTIMETRES FOR EACH OF THE LAST 10 WEEKS OF THE PREGNANCIES STUDIED.

		DURATION OF PREGNANCY IN COMPLETED WEEKS										
		30	31	32	33	34	35	36	37	38	39	40
Normal Pregnancies	No.	19	20	19	20	26	18	35	28	27	17	8
	Mean	3409	3692	4102	4655	4633	5125	5331	5657	5892	6076	5910
	+1 S.D.	3976	4370	4914	5445	5398	5942	5985	6528	6760	7003	6876
	-1 S.D.	2842	3014	3290	3865	3868	4308	4677	4786	5024	5149	4944
'Light-for-dates' Pregnancies	No.	4	7		6		7	6	6	5	4	
	Mean	2857	3044		3587		4002	4304	4181	4261	4577	
	+1 S.D.	3653	3793		4294		4778	5119	4854	5086	5573	
	-1 S.D.	2061	2295		2880		3226	3489	3508	3436	3581	

(Only one reading was obtained from 'light-for-dates' pregnancies at 32 and 34 weeks, and none at the 40th week).

Figure 19

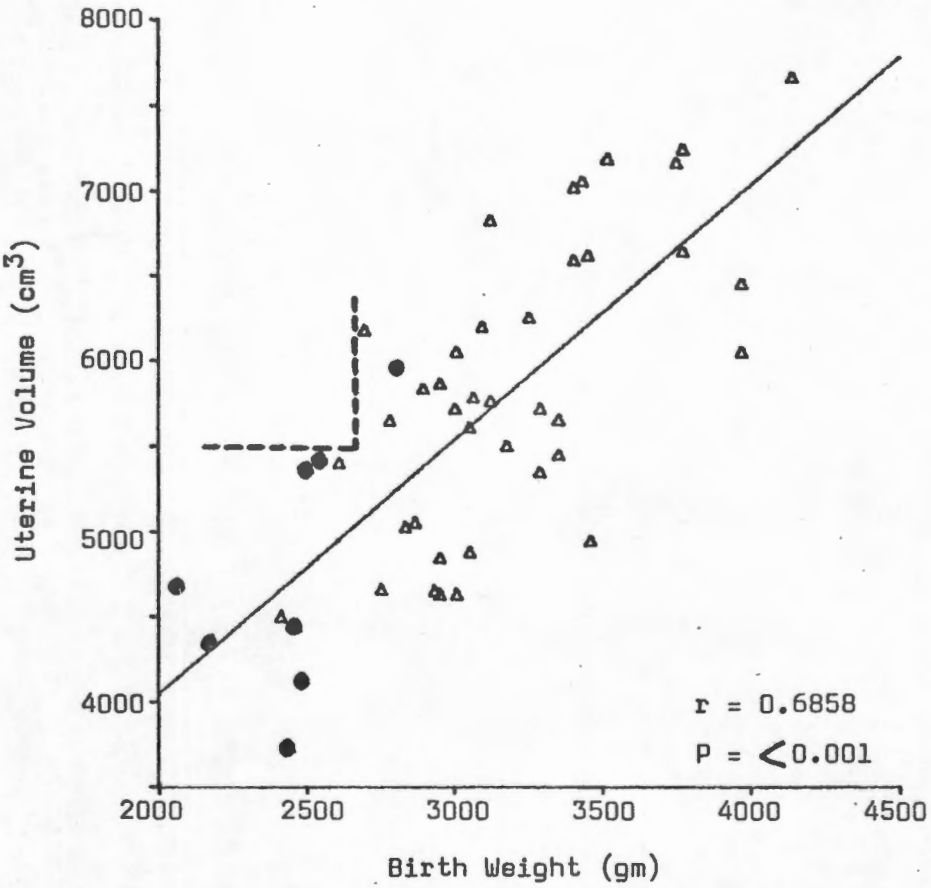


Mean and standard deviations of uterine volume for each week of pregnancy after 30 weeks.

Normal infants

'Light-for-dates' infants

Figure 20



Birth weight against the last uterine volume measurement made prior to delivery

△ Normal infants

● 'Light-for-dates' infants

possible. On only one occasion did a patient object to the examination on the grounds that it was painful. The cause for her discomfort was found to be a florid trichomonal vaginitis. Appropriate therapy with Metronidazole 200mg t.d.s. for a week effectively treated the vaginitis and the patient did not experience discomfort again.

No attempt was made to force the internal cervical os to accept the examining finger, not only to eliminate pain but in the hope that premature labour would not be stimulated. The fact that the mean duration of pregnancy was 275 (± 9.2) days, suggests that some pregnancies at least ended a few days earlier than expected (Higgins, 1956). Whether this was as a result of repeated vaginal examinations alone is conjectural, but the author feels that they must have played some role in this occurrence.

The assessment of the state of the cervix on one occasion during each of the time group periods is shown in Table IX. These figures are graphically represented in Figure 21.

TABLE IX

The Number of Patients with Cervical Grading characteristics for each time period

Cervical Grading	Duration of Pregnancy		
	30 - 33 weeks	34 - 36 weeks	37+ weeks
Grade 1	46 (97.87%)	42 (89.4%)	19 (42.2%)
Grade 2	1 (2.13%)	5 (10.6%)	16 (35.6%)
Grade 3	-	-	9 (20.0%)
Grade 4	-	-	1 (2.2%)

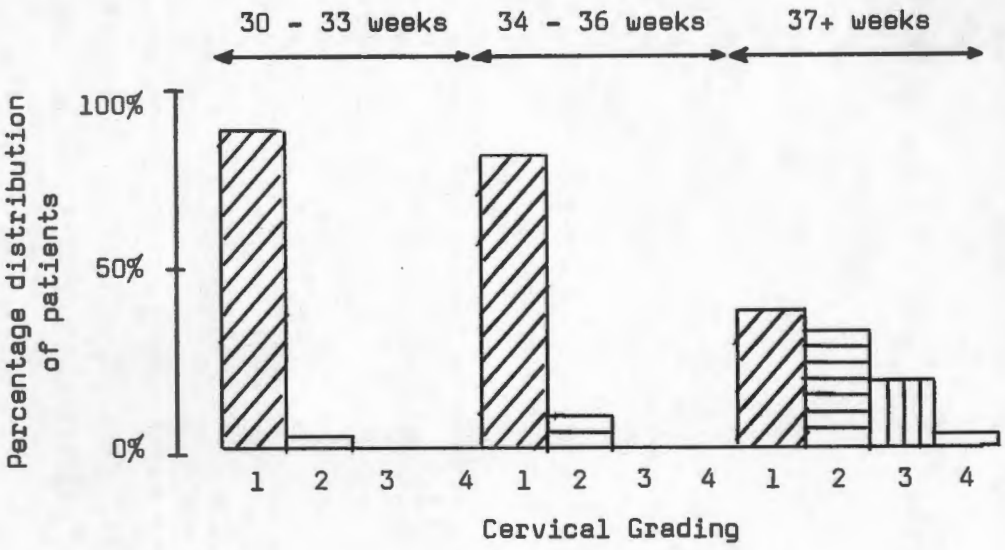


Figure 21: Graphic representation of the change in physical characteristics of the cervix with advancing gestation

The results of assessing this parameter demonstrate that although there are obvious changes in the cervical characteristics with advancing gestation, a grade 1 cervix imparts no information at all with regard to the duration of pregnancy. Conversely a grade 3 or grade 4 cervix would appear to be found only after 36 weeks, but these two grades make up less than a quarter of the total number of patients after 36 weeks.

No statistically significant difference was found between the two groups of patients i.e. those who produced normal infants and those who produced 'light-for-dates' infants.

These results are in contradistinction to those of Schaffner and Schanzer (1966) and Anderson and Turnbull (1969) but in agreement with the findings of Hendricks, Brenner and Kraus (1970). The latter authors suggest that the cervix dilates progressively in an orderly fashion during the final four weeks prior to the onset of spontaneous labour. The results from this study tend to substantiate this view.

5. ULTRASONIC CEPHALOMETRY

During the last decade ultrasonic cephalometry has assumed a role of importance in the assessment of gestational age due largely to the work of Donald in Glasgow, (Donald, MacVicar and Brown, 1965; Donald and Brown, 1961; Willocks, Donald, Duggan and Day, 1964; Donald, 1965; Willocks, Donald, Campbell and Dunsmore, 1967; and Donald, 1968). The growth of the brain, expressed as changes in weight, volume and diameter, appears to be less affected than body weight and length by changes in the intra-uterine conditions (Gruenwald, 1968) and could, therefore, be expected to be a reliable index of gestational age.

Before the advent of reliable methods of paediatric assessment of gestational age, the diagnosis of prematurity rested solely upon the birth weight of the infant. In an attempt to diagnose prematurity by this definition prior to delivery, a number of workers tried to relate the biparietal measurement to the weight of the baby (Taylor, Holmes, Thompson and Gottesfeld, 1964; Anderson and Niswonger, 1965). At best they were incorrect by 500 grams and it was Willocks et al (1964) who demonstrated an increase in biparietal diameter from the 20th to the 40th weeks of pregnancy. This finding was confirmed by Thompson et al (1965) and Hibbard and Anderson (1967).

More recently Campbell (1969) reported that from the 20th to the 30th week of pregnancy, the biparietal diameter values were confined within narrow limits for a certain week of gestation, and that measurement during this period enabled him to predict the gestational age to within ± 8.4 days in 95% of cases. Willocks (1962) reported a mean growth rate of 1.5mm per week during the last 10 weeks of pregnancy and noted that the mean for the period 30 to 36 weeks was higher than that for the period 36 to 40 weeks. Campbell (1969)

found a growth rate of 2.8mm per week between 20 and 30 weeks and the same rate of 1.5mm per week thereafter but noted a much wider scatter about the mean during the last 10 weeks of pregnancy. These figures suggest that factors affecting intra-uterine growth may have less influence on the developing nervous system before the 30th week of pregnancy than thereafter.

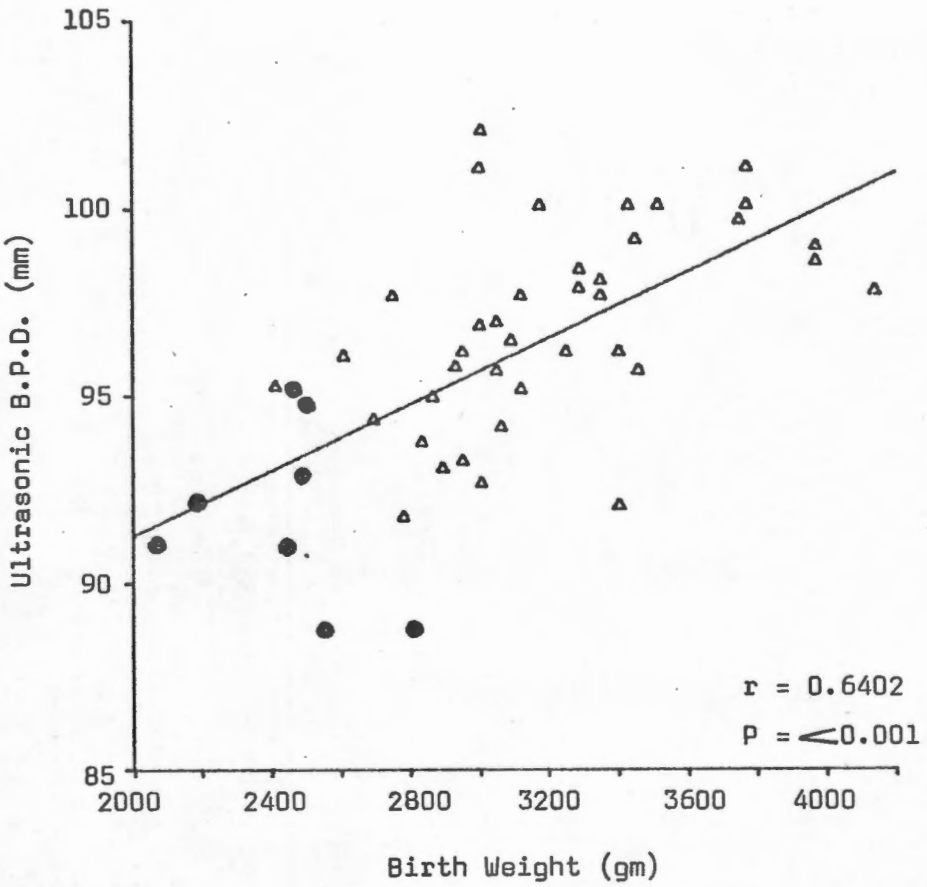
In attempting to assess the relationship between ultrasonic biparietal diameter measurements and birth weight a problem became immediately apparent. Although some deliveries occurred within a few days of the last measurement being made, occasionally this time interval was over three weeks. In spite of this the last measurement obtained was plotted against the birth weight and the results are seen in Figure 22. The very wide scatter may in part be due to those infants who continued growing at normal rates for some considerable time after the last measurement was taken. Nevertheless, the impression gained is that the absolute measurement of the biparietal diameter as measured ultrasonically is unable to assist in the prediction of birth weight.

Correlation of the biparietal diameter measurement with gestational age was more rewarding. Figure 23 shows this plotted graphically in those pregnancies resulting in normal infants. Ninety-five percent confidence limits have been plotted and it can be seen that in general, a good correlation exists between the two. When considering the individual, however, it can be seen that the range within 95% confidence limits at any particular stage of pregnancy after 30 weeks is 10mm. Similarly the range in time for any single measurement is just over 5 weeks.

Segmental analysis reveals that a measurement of more than 92mm indicates a gestational age of more than 34 weeks, and a

measurement of less than 90mm indicates a gestational age of less than 36 weeks. This segmental analysis is only valid provided that the pregnancies are normal as can be seen from Figures 25 and 26, where the biparietal diameter measurements were plotted against gestational age in both race groups independently. It is obvious that there is no statistically significant difference between the two race groups but that the 'light-for-dates' infants have significantly smaller measurements than their normal counterparts.

Figure 22

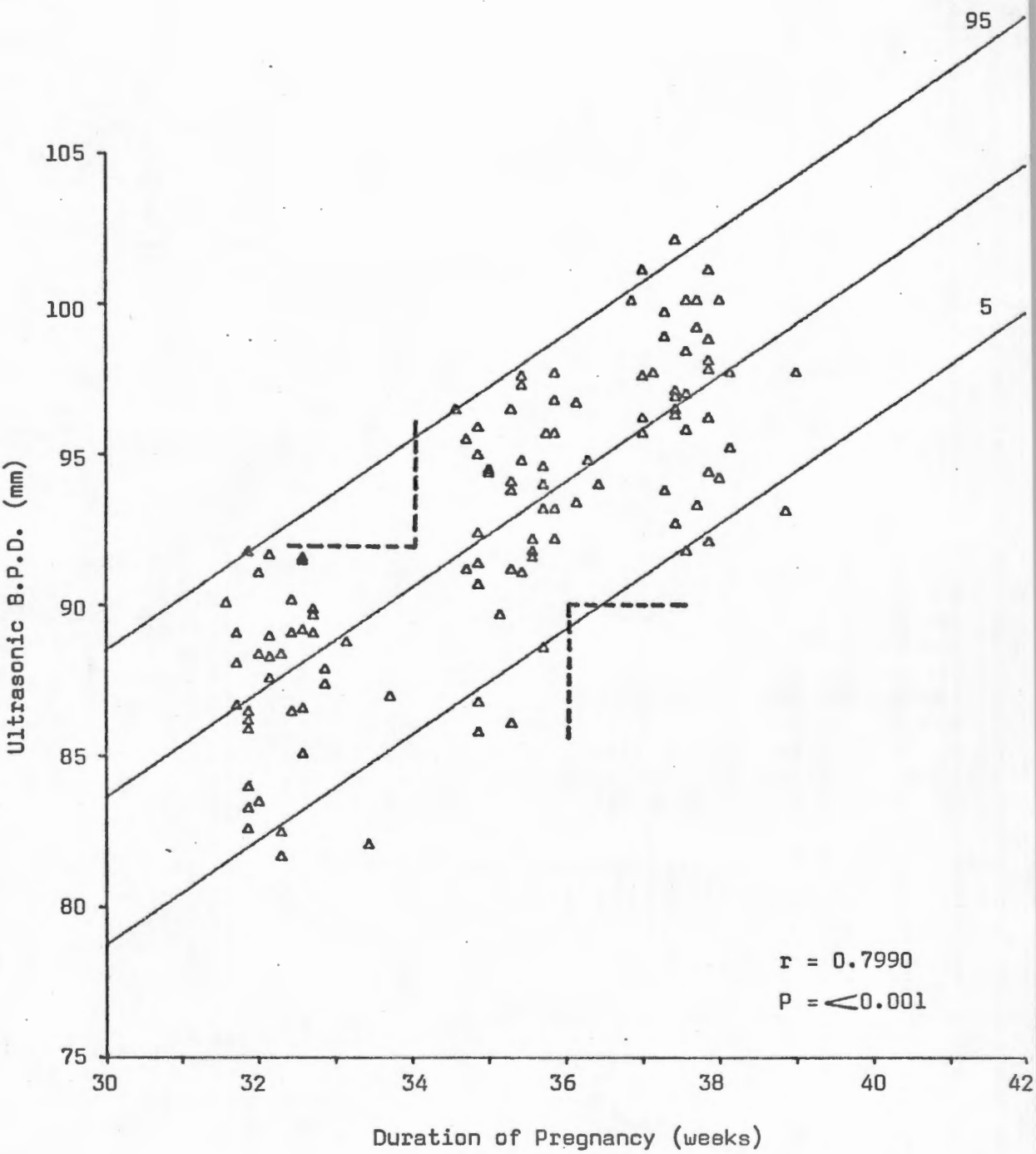


The relationship between the last ultrasonic biparietal diameter measurement and the birth weight.

△ Normal infants

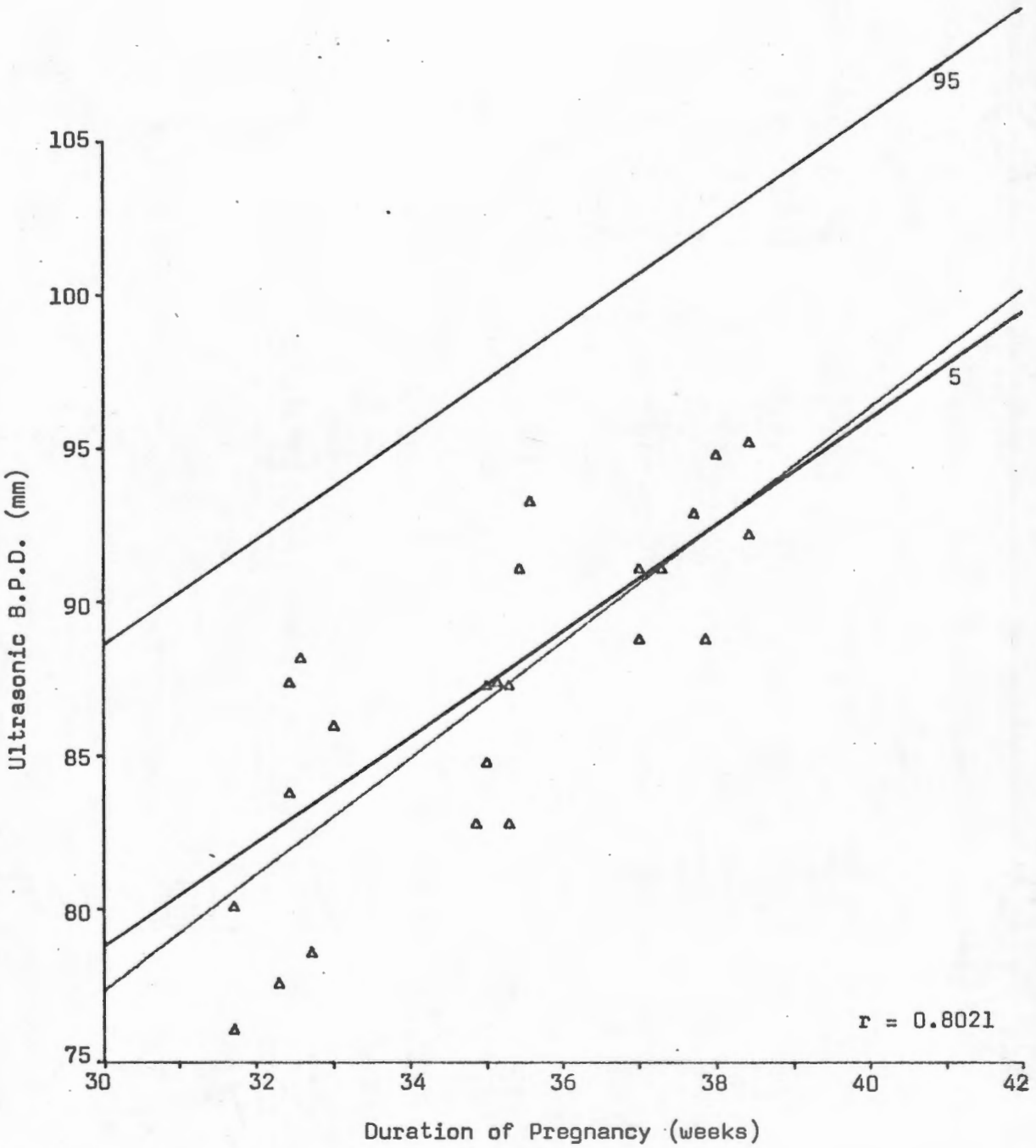
● 'Light-for-dates' infants

Figure 23



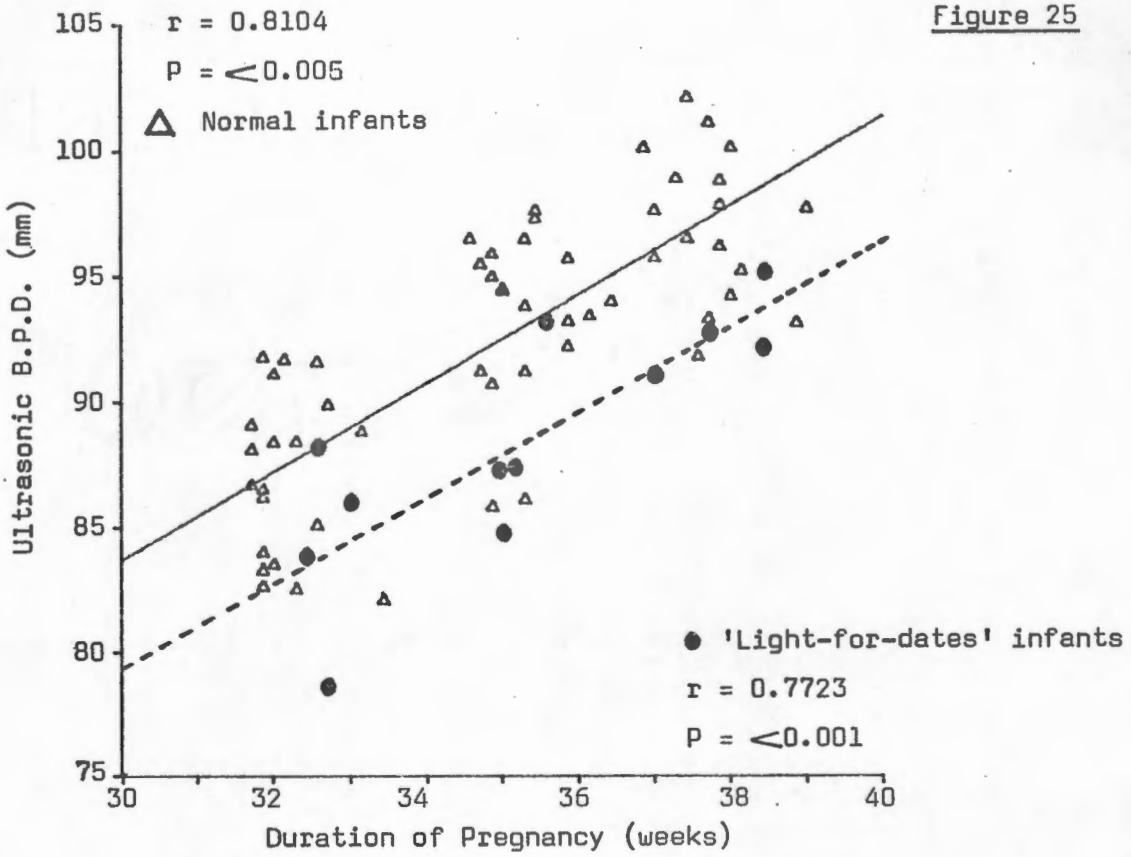
Ultrasonic biparietal diameter measurements plotted against gestational age in those patients producing 'normal' infants.

Figure 24



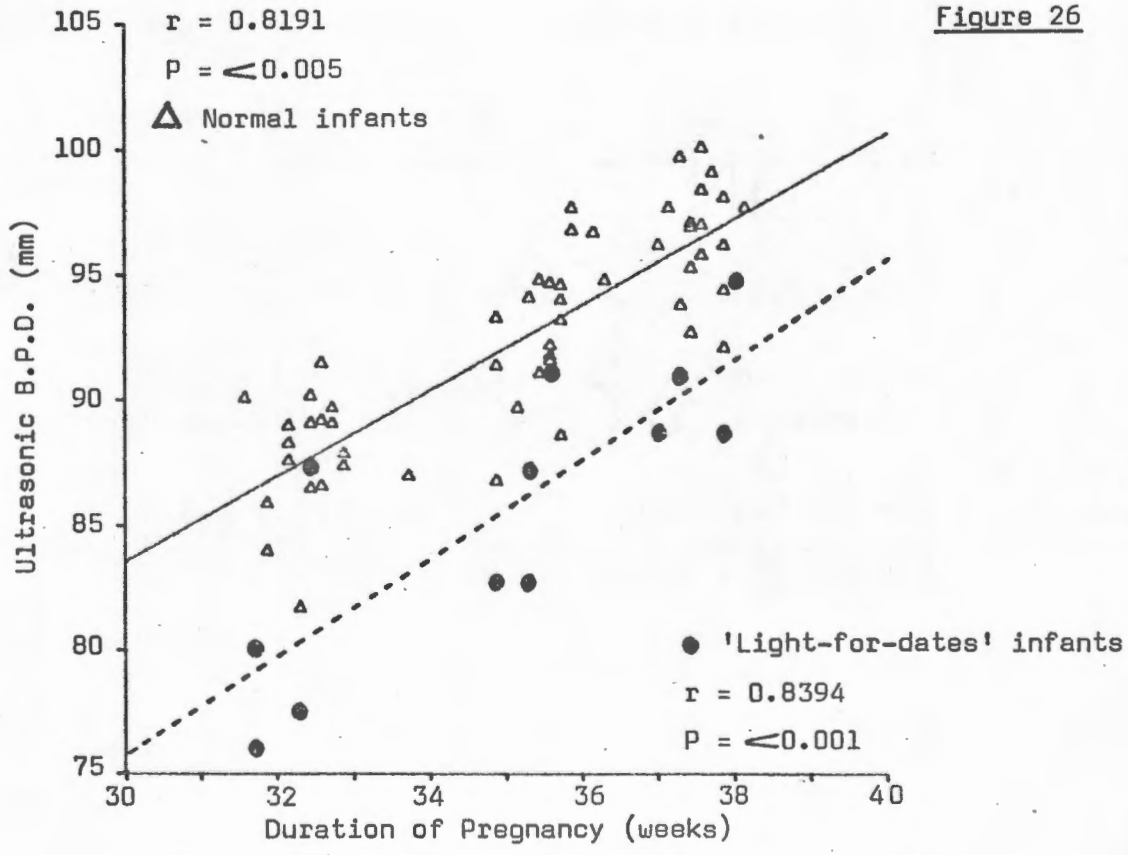
Ultrasonic biparietal diameter measurements plotted against gestational age in those patients delivered of 'light-for-dates' infants. The 95% confidence limits are those calculated for Figure 23.

Figure 25



Ultrasonic B.P.D. against gestational age in European patients

Figure 26



Ultrasonic B.P.D. against gestational age in Non-European patients.

6. LIQUOR CYTOLOGY

A. History

One of the first reports concerning the cellular component of the amniotic fluid came from Bourgeois (1942), who described the fetal squamous cell for the first time. He attempted to elucidate the problem of diagnosis of premature rupture of the membranes and concluded that the fetal squame was a polygonal, translucent degenerated cell which had its origin in the vernix caseosa and was seldom seen prior to 32 weeks gestation. Shettles (1956) in studying the sex chromatin of cells from liquor amnii suggested that these cells had their origin in either the amnion or the fetus - skin, alimentary, respiratory or urinary tracts, and the umbilical cord. Danforth and Hull (1958) examined the so-called amnion cells closely and from their observations postulated that these cells were unique in structure and performed a secretory function.

Nile Blue sulphate was first introduced to liquor cytology by Kittrich (1963) when he found that cells of fetal origin stained orange whereas maternal cells stained blue, providing a simple and accurate means of diagnosing premature rupture of the membranes. Liquor amnii obtained by amniocentesis was examined by Van Leeuwen et al (1965) who concluded that the fetal cells had their origin in the vernix caseosa.

Brosens and Gordon (1965, 1966) introduced the concept of a progressive change in the cellular compartment of amniotic fluid with increasing gestational age, when, using the Nile Blue stain, they recorded an increase in the percentage of orange-staining fat cells towards term. They contended that these cells had their origin in the sebacious glands of the fetal skin and their increase

was an index of maturity of fetal skin. These same authors (1967) suggested a sudden rise in fat cell percentage after 36 weeks gestation. Following these classical reports, other authors (Bishop and Corson, 1968; Anderson and Griffiths, 1968; Sharma and Trussell, 1970) confirmed the finding of 10% or more fat cells after 36 weeks gestation. Subsequent reports have shown, however, that although there is a rise in the fat cell count towards term, up to 40% of term gestations have been found to have less than 10% of fat cells (Sharp, 1968; Chan et al, 1969; Barnett and Nevin, 1970; Lind, 1970). Stenbäck and Ojala (1969) concluded that histochemical properties of liquor cells were of little value in estimating gestational age. Sharp (1968), Huisjes, (1968), Parmley and Miller (1969) and Bennett and Morris (1972) have all questioned the origin of the 'fat' cell originating from the sebaceous gland by virtue of the holocrine nature of this organ.

Huisjes (1970) and Huisjes and Arendzen (1970) concluded that the fat cell of Brosens and Gordon (1965) was the same cell as their polygonal cell using the Harris-Shorr stain. Different authors (Anderson and Griffiths, 1968; Sharp, 1968; Sharma and Trussell, 1970; Huisjes and Arendzen, 1970) have found that various pathological states of pregnancy as well as degrees of intra-uterine malnutrition have not affected the fat cell count.

In the cytological assessment of gestational age, emphasis has recently been placed on a differential cell count like that used in vaginal cytology (Votta et al, 1968; Mandelbaum and Evans, 1969; Bishop and Corson, 1968; Huisjes, 1970; Bishop and Pollock, 1970; Huisjes and Arendzen, 1970; Lind, 1970). This method has its basis on the assumption that the fetal squamous cells originate in the fetal skin and are increasingly desquamated with advancing

maturity (Floyd et al, 1969; Votta et al, 1968).

B. Nile Blue Stain

On examination of the slides stained with Nile Blue, it became apparent that considerable aggregation of fat cells occurred in the last three or four weeks of pregnancy. Figure 27 shows the percentage of patient samples that demonstrated clumping of fat cells after the 36th week of gestation.

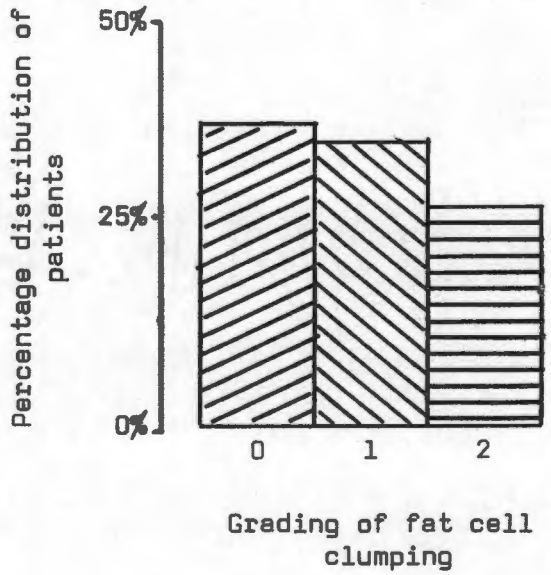
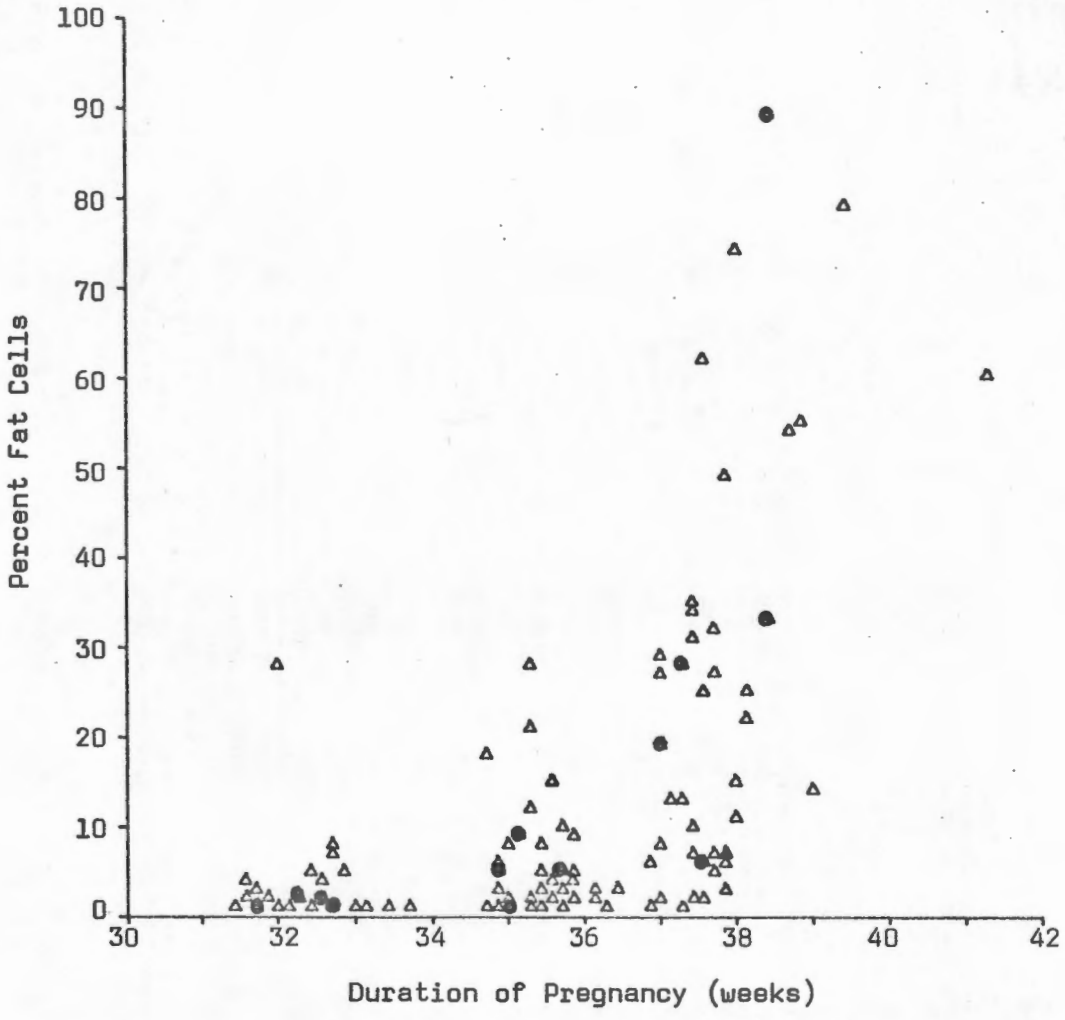


Figure 27: Graphic representation of the percentage of patient samples which exhibited different grades of clumping of fat cells after the 36th week of pregnancy.

The percentage of fat cells was found to show a rapid rise when plotted against gestational age, but this rise began earlier than the 36 week point of Brosens and Gordon. From Figure 28 it will be seen that a number of cases have a count in excess of 10% prior to 36 weeks and a much larger number a count of less than 10% after 36 weeks. One patient had a count of

Figure 28



The percentage of fat cells found in the liquor amnii during the last 10 weeks of pregnancy.

△ Normal infants

● 'Light-for-dates' infants

(To facilitate plotting of the above figure, all readings of 0% were omitted).

28% at 32 weeks and successive counts in this case were 28% and 29% respectively. No explanation was found for these readings being so high at 32 and 35 weeks.

Infants classified as 'light-for-dates' did not appear to differ from the normal ones in respect of their fat cell counts.

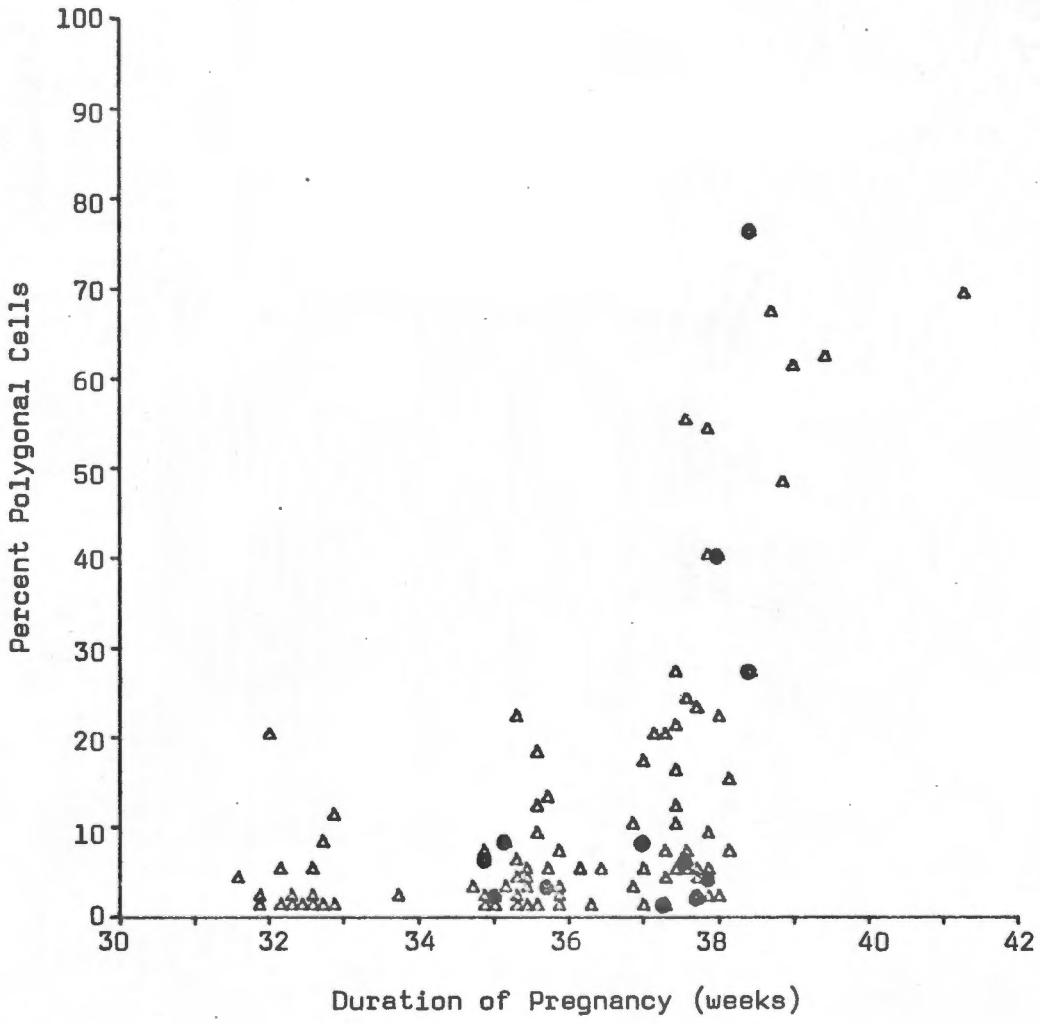
C. Harris-Shorr Stain

Assuming that Huisjes (1970) is correct in maintaining that the fat cell and the polygonal cell are the same cell, a similar type of rise in polygonal cells relative to gestational age could be expected. When the results of the differential counts were examined, each cell type was correlated with gestational age. Figure 29 shows a rise in polygonal cells very similar to that of the fat cells (Figure 28). Once again there was no difference between the counts obtained from the normal infants and those obtained from the 'light-for-dates' infants.

Figure 30 shows the correlation between the two types of cell to be a highly significant one ($P = < 0.001$). Why the fat cell counts tend to be higher than the polygonal cell counts for the same patient at the same period of gestation is difficult to explain. The author feels that the explanation may lie in the preparation of the smear viz. the fat cells are counted in uncentrifuged liquor, whilst the Harris-Shorr stain is applied to a smear made from a centrifuged mass of cells. This leads to clustering of cells and extreme difficulty in counting unless the cells are in layers one cell thick. Nevertheless the author feels there is sufficient evidence to suggest that these two cells are, in fact, one and the same cell.

Floyd et al, (1969) and Lind (1970) suggest a predominance of precornified and cornified cells after 36 weeks gestation with

Figure 29



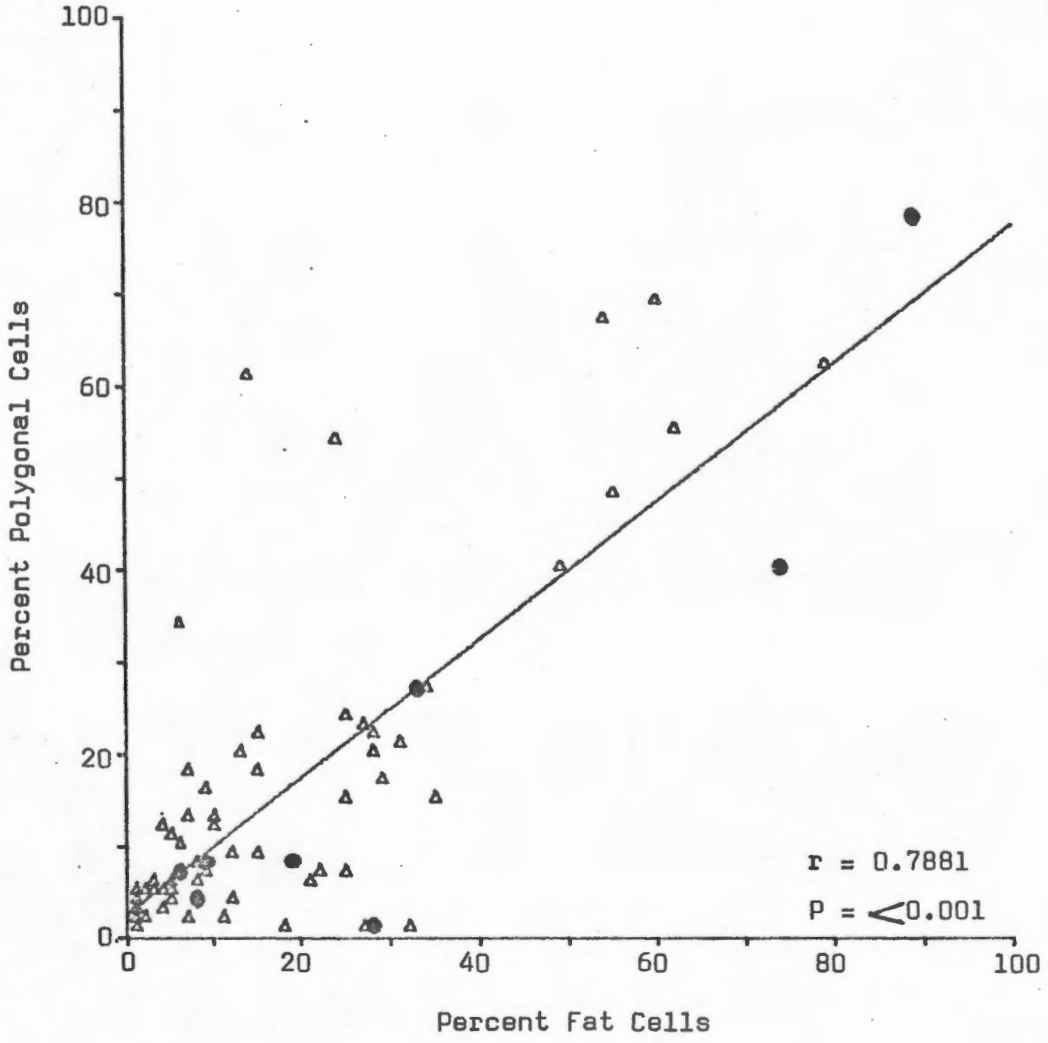
The percentage of polygonal cells found in liquor amnii during the last 10 weeks of pregnancy.

△ Normal infants

● 'Light-for-dates' infants

(To facilitate plotting of the above figure, all readings of 0% were omitted).

Figure 30



The relationship between fat and polygonal cells in the liquor amnii at equal periods of gestation. For the sake of clarity, duplicate results and those of 0% have been omitted.

△ Normal infants

● 'Light-for-dates' infants

a concomitant decrease in parabasal and basal cells. Neither make any mention of the polygonal cell.

Examination of the results of this series of patients reveals a very low basal cell count from 31 weeks onwards with a marginal decrease i.e. 2.1 (± 1.8)% at 30 - 33 weeks to 1.3 (± 0.6)% at 37+ weeks. Contrary to the findings of the above authors, the intermediate, superficial and anucleate squamous cell counts all decreased with advancing age, (Figures 31, 32 and 33). The very large variance of the results makes prediction of gestational age impossible in the vast majority of cases. Counts of less than 10% tend to suggest a gestational age of more than 35 - 36 weeks.

The explanation for the drop in the precornified and cornified elements in contradistinction to that found by other workers, is the inclusion of the polygonal cell in the total cell count. If the latter rises sharply after 37 weeks then a corresponding fall in the relative proportions of the other cell types must occur.

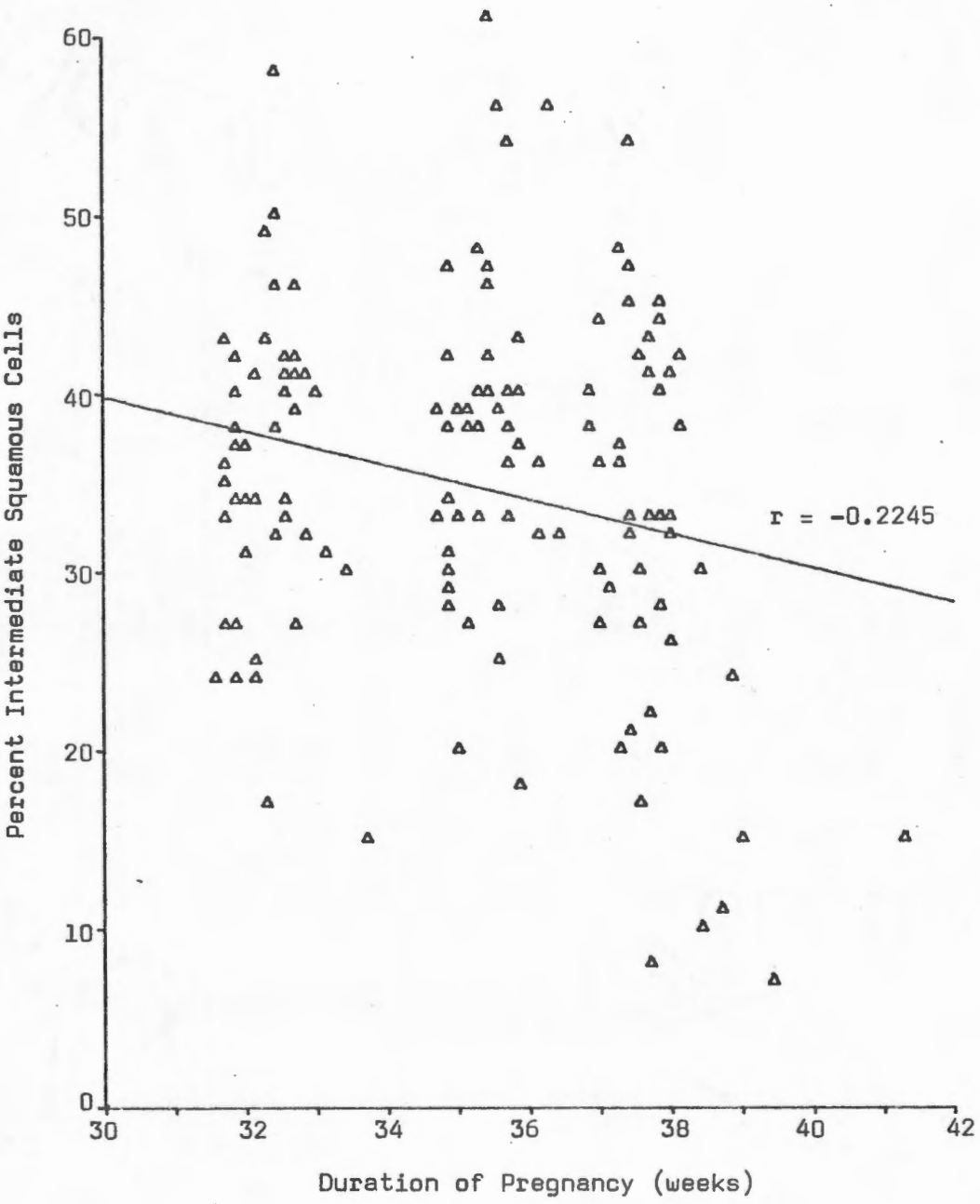
Amnion cells (Wachtel et al, 1969) were rarely seen and bore no relationship to gestational age. This finding is in agreement with Floyd et al (1969) and Huisjes (1970).

As with the polygonal cell counts, 'light-for-dates' infants exhibited no differences from the normal infants with respect to these latter cell types.

D. <u>The Influence of Sex upon the Cytological Population of Liquor Amnii</u>.

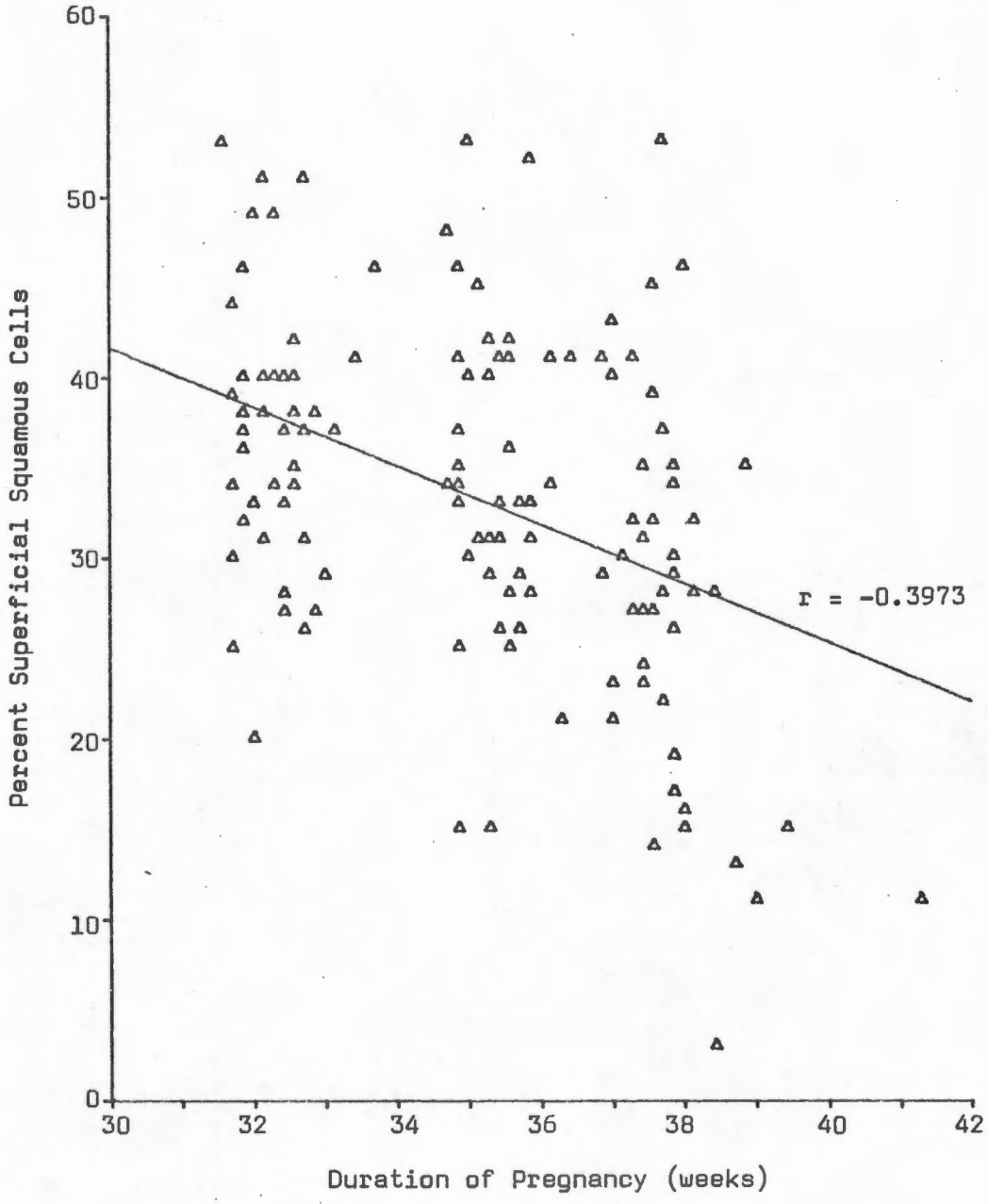
Using the Harris-Shorr stain, it became apparent that there were two different types of smear obtained when the liquor amnii cell deposit was examined microscopically. This difference

Figure 31



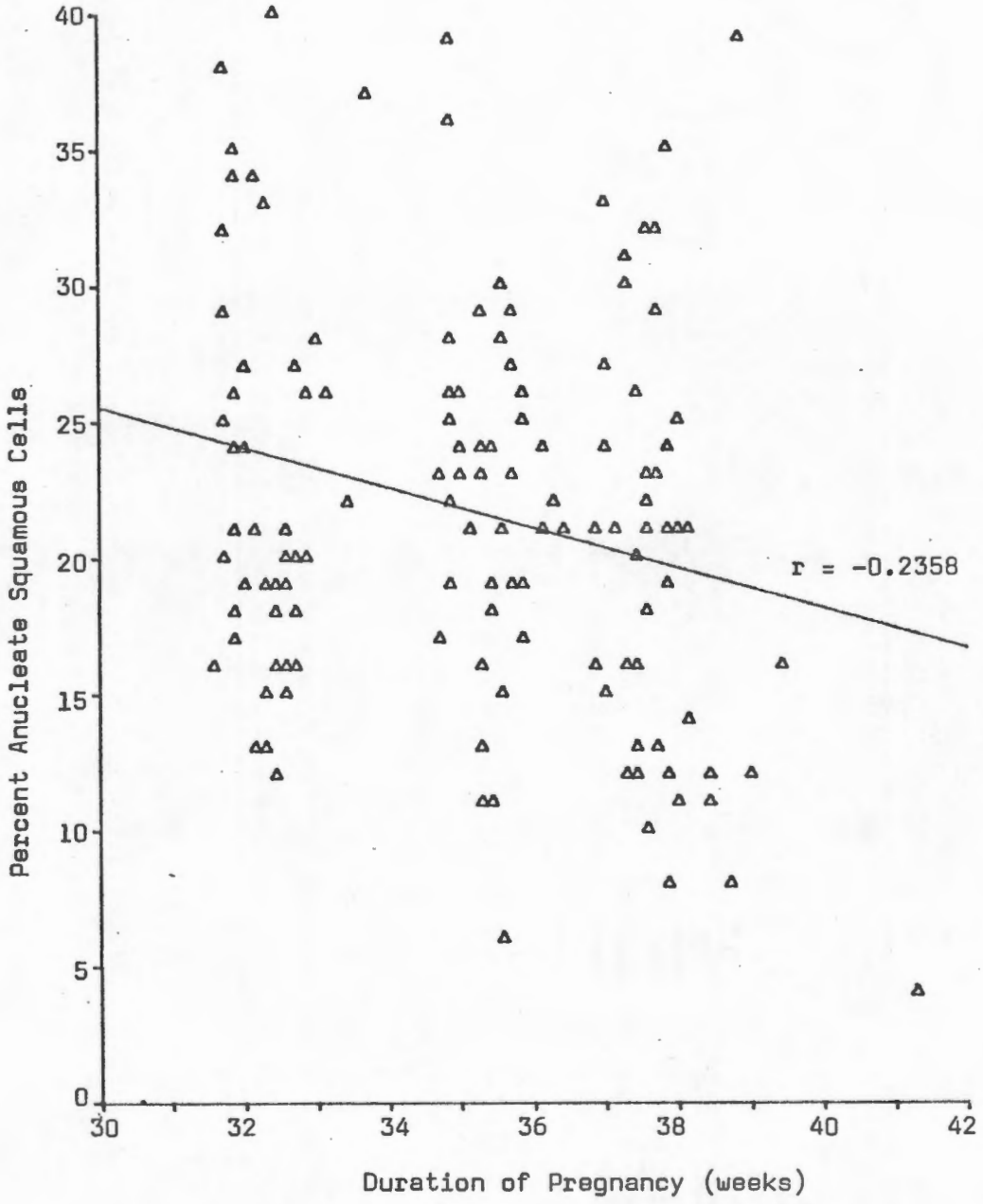
The percentage of intermediate squamous cells found in the liquor amnii during the last 10 weeks of pregnancy.

Figure 32



The percentage of superficial squamous cells found in the liquor amnii during the last 10 weeks of pregnancy.

Figure 33



The percentage of anucleate squamous cells found in the liquor amnii during the last 10 weeks of pregnancy.

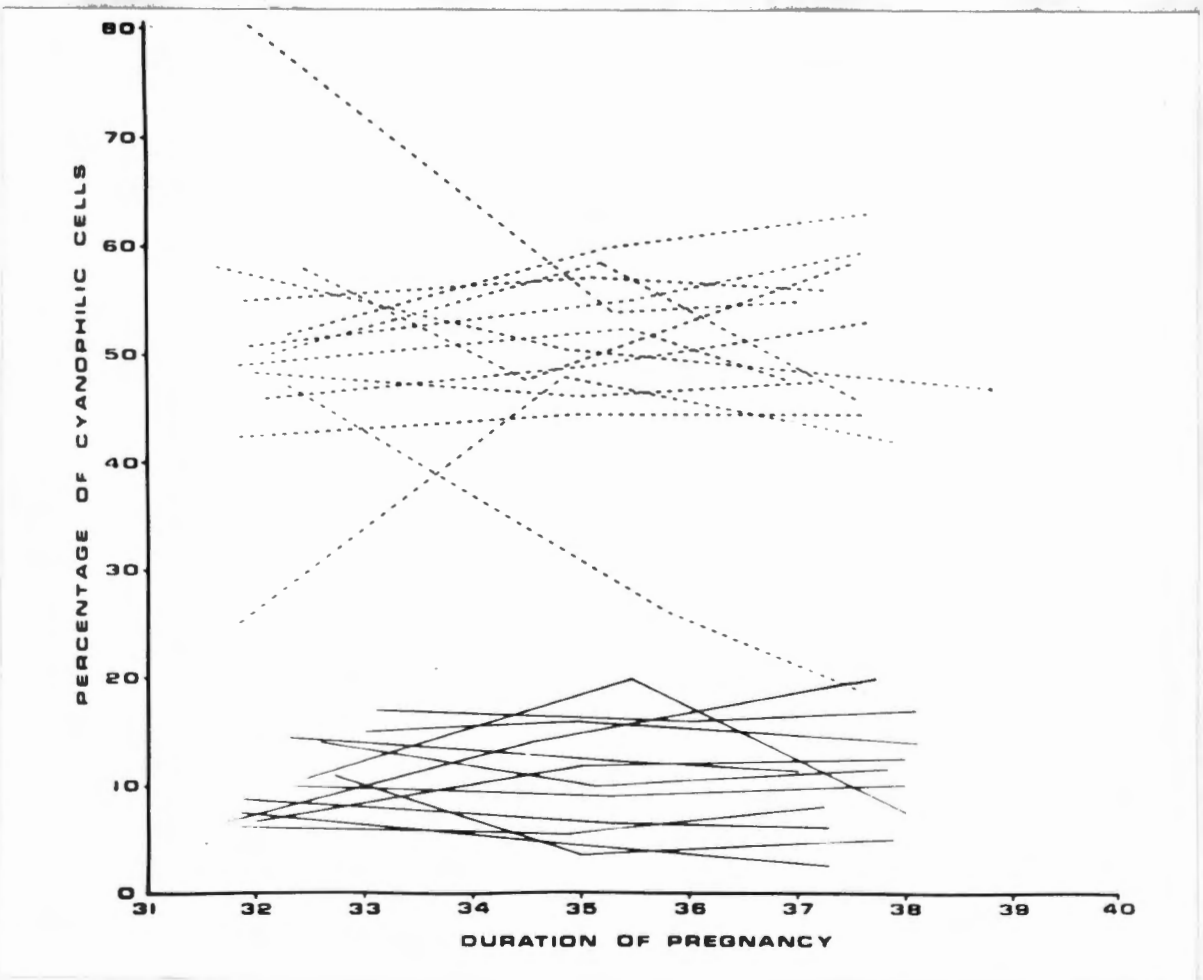
was found to be different proportions of cells staining cyanophilic and eosinophilic respectively i.e. some smears appeared to contain a higher percentage of cyanophilic staining cells than did others. This difference appeared to have little relationship to the duration of pregnancy and was not specifically confined to one particular morphological type of cell, although the highest proportion of cyanophilic staining cells appeared to be intermediate in type.

A search was made for the explanation for the differences in staining characteristics and a prospective study was made of the last 25 patients to deliver in the study group. Five hundred cells were counted on each slide and the percentage of eosinophilic and cyanophilic squamous cells determined. Following delivery of these patients, it was found that the sex of the infant was the only factor which correlated well with the percentage of cyanophilic cells (Bennett, Morris and Davey, 1972).

From Figure 34 it can be seen that when there were more than 25% of cyanophilic cells in the cell population the infant was female in 100% of cases. When the count was less than 20%, the chances of the infant being male were 94%. The overlap of the two groups was attributable to a single case which showed a drop from an initial figure of 47.2% at 32 weeks. This infant was re-examined 9 weeks after delivery and a sterile throat swab could not be introduced into the vagina. A buccal smear taken at this time was mislaid by the laboratory. It was unfortunately decided to defer further investigation and when it became apparent that a second buccal smear had to be taken, the family were found to have moved from their previous address and could not be traced.

Barr and Bertram (1949) were the first to describe the

Figure 34



Cyanophilic cell counts in liquor amnii during the last 10 weeks of pregnancy in 25 cases

The lines (- - - -)(————) indicate serial estimations in individual patients.

- - - - - - female infants

———— male infants

presence of sex chromatin in the nuclei of cells taken from female cats and then confirmed their findings using human cells. James (1956) and Dewhurst (1956) were the first to describe the presence of the sex chromatin or Barr body in the cells found in liquor amnii. Dewhurst found that where the fetus was male, this figure was less than 5%. The accuracy of prenatal sex determination by this method was confirmed by Fuchs and Riis (1956); Makowski, Prem and Kaiser (1956); Serr, Sachs and Danon (1956); Shettles (1956) and Keymer, Silva-Inzunza and Coutts (1957). It was found that this method of antenatal sex determination was accurate from early in the second trimester onwards (Serr, Sachs and Danon, 1957).

The obvious application of this work was the antenatal diagnosis of sex-linked diseases. Riis and Fuchs (1960) made the correct prediction of sex in two women known to be haemophilia carriers and Serr and Margolis (1964) correctly predicted the sex of a fetus carried by a woman who had had three sons die of Werdnig-Hoffman progressive spinal muscular atrophy. Abortion was refused by the patient and her fourth son died at the age of 14 months.

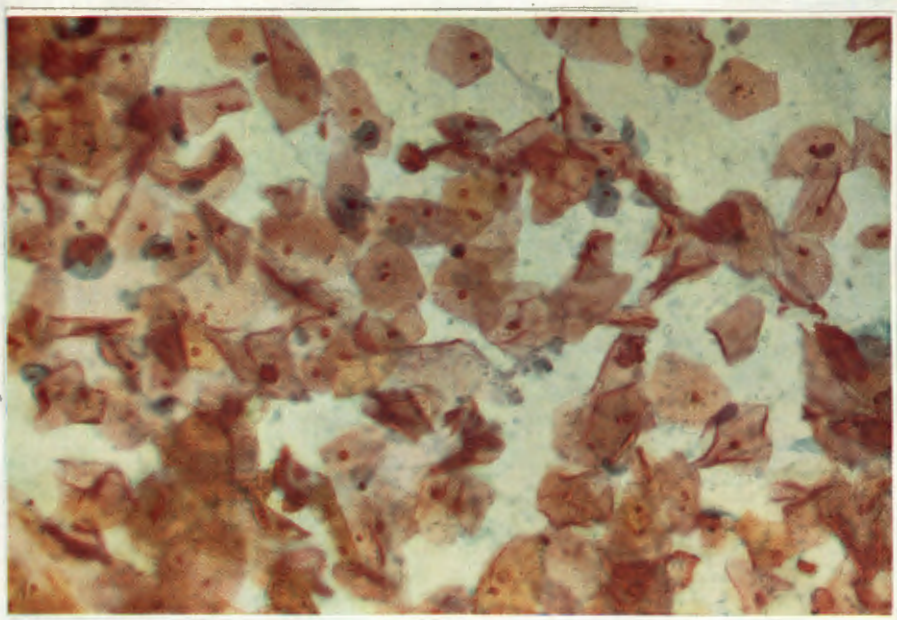
Rosa and Fanard (1949) found the liquor from male infants when stained by the Papanicolaou method contained fewer blue staining cells than did liquor from female infants. Huisjes (1968a; 1968b), using the Harris-Shorr stain, concluded that the vast majority of these cyanophilic staining cells found in the liquor amnii of female infants were intermediate in type and of vaginal origin.

Bennett and Morris (1972), in a study into the origin of the cells found in the liquor amnii, found that the exclusive sites of origin of the intermediate type of cell were the buccal

mucosa and the vaginal vestibule. This would account for the presence of such cells in liquor from male infants, although only infants born after 36 weeks were available to them for study.

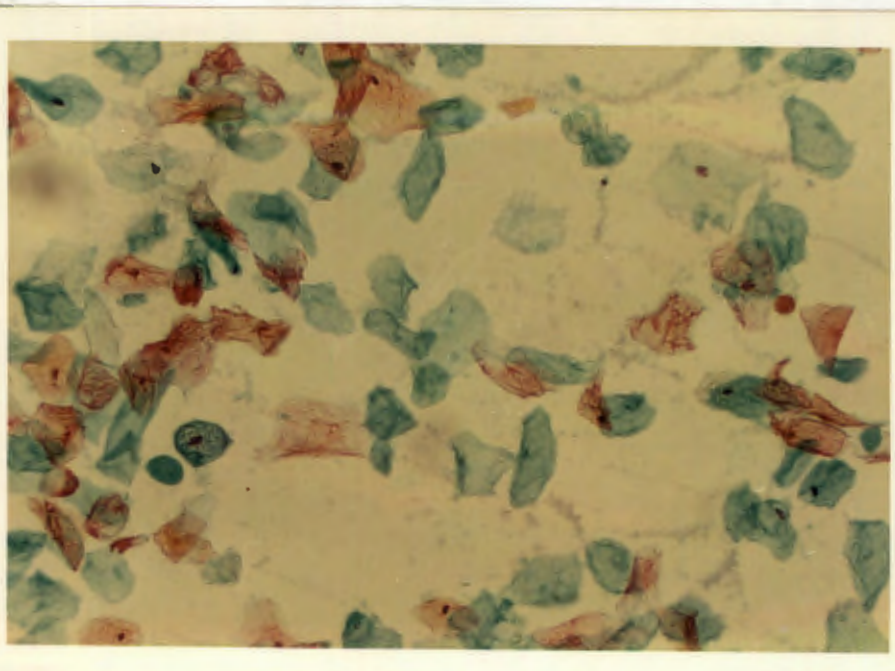
Figures 35 and 36 show the difference in the staining characteristics of cells from the liquor amnii of male and female infants with regard to the percentage of cyanophilic staining cells present.

Figure 35



Cells in the Liquor Amnii of a male fetus at 37 weeks gestation showing few cyanophilic cells (x 200).

Figure 36



Cells in the Liquor Amnii of a female fetus at 36 weeks gestation showing a higher percentage of cyanophilic cells (x 200)

7. LIQUOR BIOCHEMISTRY

A. History

Hippocrates is accredited with the belief that the main source of liquor is fetal urine (Shaw and Marriott, 1949; Plentl, 1966) and Joseph Needham, a 17th century man-midwife described the liquor as a 'private pond' for the fetus (Holland, 1959). Harvey in 1651 suggested fetal swallowing as the means of removal of liquor amnii (Plentl, 1966). On the basis of a lowered osmotic pressure in the amniotic fluid, Zangemeister and Meissel in 1903 (Harbert et al, 1968) concluded that dilution with some hypotonic solution was the only explanation and Uyeno (1919) suggested the presence of fetal urine in the liquor amnii. Evidence of fetal swallowing was shown by Mislocki in 1921 and Vrbitch in 1924 (Harbert et al, 1968). Taussig (1927) wrote "there can be no reasonable doubt that the source of the amniotic fluid is the amnion itself and that the theory from the days of Hippocrates to the present century, that this fluid merely consists of fetal urine is unfounded", but had no evidence for this statement.

The first recorded attempts at volumetric measurements of amniotic fluid by means of amniocentesis were those of Dieckmann and Davis (1933) but their results were very varied and no mention is made of gestational age or pathological status of the pregnancy.

A large lacuna exists in medical literature regarding the liquor amnii from then onwards and it is only in the last 15 years, with the acceptance of amniocentesis as a practical procedure, that the physiology of liquor amnii has been studied.

Direct volumetric measurement following hysterotomy enabled Gillibrand (1969) and Lind and Hytten (1970) to show a close

correlation between the volume of liquor amnii and fetal weight as well as gestational age.

Dye dilution techniques have allowed various authors to calculate the volume of liquor amnii during the second half of pregnancy (Hutchinson et al, 1955; Elliot and Inman, 1961; Charles et al, 1965; Charles and Jacoby, 1966; Marsden and Huntingford, 1965; Gadd, 1966; Gillibrand, 1969; Andrews, 1970; Normington, 1970). Unlike in the first half of pregnancy, there appears to be no correlation whatsoever between liquor volume and gestational age during the second 20 weeks. Very large ranges, sometimes more than 1000ml, have been reported at the same weeks of gestation and the consensus of opinion would appear to be that the volume rises to a maximum at 37 - 38 weeks and then declines to term. The decrease in volume after 40 weeks appears to be even more marked (Hytten and Leitch, 1971).

At least 20 theories have been proposed to explain the formation of amniotic fluid, but only a few are now acceptable (Wagner et al, 1968). Amniotic fluid is present in early pregnancy before the development of fetal tissues and, in fact, may be found within the amniotic cavity of a blighted ovum in which the fetus is absent or rudimentary (Jeffcoate and Scott, 1959). Thus it would seem that since the cells of the amnion have secretory characteristics (Lister, 1968), fluid would initially be formed by the amniotic membranes. Similarly the fetal dermal membrane and germinativum probably offer little resistance to the passage of water and solutes (Lind, 1969; Parmley and Seeds, 1970) until ± 17 weeks when keratinization begins. The conclusion is, therefore, that the amniotic fluid would appear to be a dialysate of both maternal and fetal serum as initially proposed by Makepeace

et al (1931), as well as an extension of the fetal extra-cellular fluid (Hytten and Leitch, 1971). This theory is suggested by osmolatity estimations and biochemical analyses of maternal and fetal serum as well as liquor amnii (Lind, 1969; Lind et al, 1969).

Early authors assumed the amniotic fluid to have a relatively stable composition and published average values for the constituents (Zangemeister and Meissel, 1903; Uyeno, 1919; Taussig, 1927; Acosta-Sison, 1936). It is surprising to find recent authors perpetuating this misconception (Hannon, 1959; Bonsnes, 1966).

Many biochemical studies (Chez et al, 1964; Plentl, 1966; Scarpelli, 1967; Seeds and Hellegers, 1968; Mandelbaum and Evans, 1969; Miles and Pearson, 1969; Gillibrand, 1969; Begneaud et al, 1969; Cherry and Dolger, 1969; Lind et al, 1969; Mattison, 1970; Roopnarinesingh, 1970; Doran et al, 1970; Biggs and Duncan, 1970) have led to the conclusion that contributions to the liquor amnii are made by the fetal respiratory tract, salivary glands and kidneys, as well as the umbilical cord (Wagner et al, 1968). Specific biochemical changes occurring with advancing gestation are discussed later.

The disposal of liquor amnii, like its formation, has been the subject of conjecture for many years. The complete answer to this problem has yet to be found but it would seem likely that deglutition by the fetus plays the dominant role (Hytten and Leitch, 1971) as originally proposed by Harvey in 1651 (Plentl, 1966). By using amniographic techniques, McLain (1963) was able to prove the theory of intra-uterine swallowing by the fetus. Less argument exists regarding the volume of liquor swallowed, and Pritchard

(1965) claimed that ± 450ml per day was swallowed by a term fetus.

Jeffcoate and Scott (1959) postulated a deficient swallowing mechanism to account for the common association of anencephaly and polyhydramnios. They also suggested that there was a causal relationship between urinary tract malformations and the finding of oligohydramnios i.e. either failure of the urinary tract to produce urine or failure of communication between the urinary tract and the amniotic fluid. Gadd (1966) suggested that atresias of the gastrointestinal tract and malformations of the central nervous system resulting in a deficient or defective deglutition centre were the cause of the majority of instances of polyhydramnios.

The very large exchange rates of water reported by Hutchinson et al (1955) and Harbert et al (1968), are accounted for by an error in interpretation according to Haworth et al (1968). These latter authors suggest that the total turnover rate includes the exchange of molecules by Brownian movement through all surrounding membranes and that such exchange does not affect the composition of the compartment and nor does it provide a measure of the rate of formation. The production rate of 16-42ml per hour reported by Biggs and Duncan (1970) would seem to the author more realistic than the figure of 468ml/hour suggested by Hutchinson et al (1955). This is pertinent to the finding of Chez et al (1964) that the Rhesus monkey fetus excretes 5ml of urine per kilogram per hour, although it is admitted that extrapolation to the human fetus is not necessarily possible.

B. Liquor Urea Concentration

Uyeno (1919) was one of the first workers to find urea in the amniotic fluid, but it was Lind (1969); Lind et al (1969),

Mandelbaum and Evans (1969); Gillibrand (1969) and Lind (1970) who related its concentration to gestational age. Gillibrand (1969) suggested a level of 31mg% at term and Lind (1969) a figure of 30mg% after 38 weeks gestation. Lind (1970) had previously suggested that the urea concentration in the liquor amnii was higher than that in the maternal serum after 30 weeks of gestation.

Figure 37 shows the very wide scatter of results obtained in this series, very similar to the results of Lind et al (1969), although higher means were obtained than reported by these authors (Appendix E, Table XX). The pregnancies resulting in 'light-for-dates' infants yielded slightly higher values than the normal pregnancies, but the differences between the means are not statistically significant.

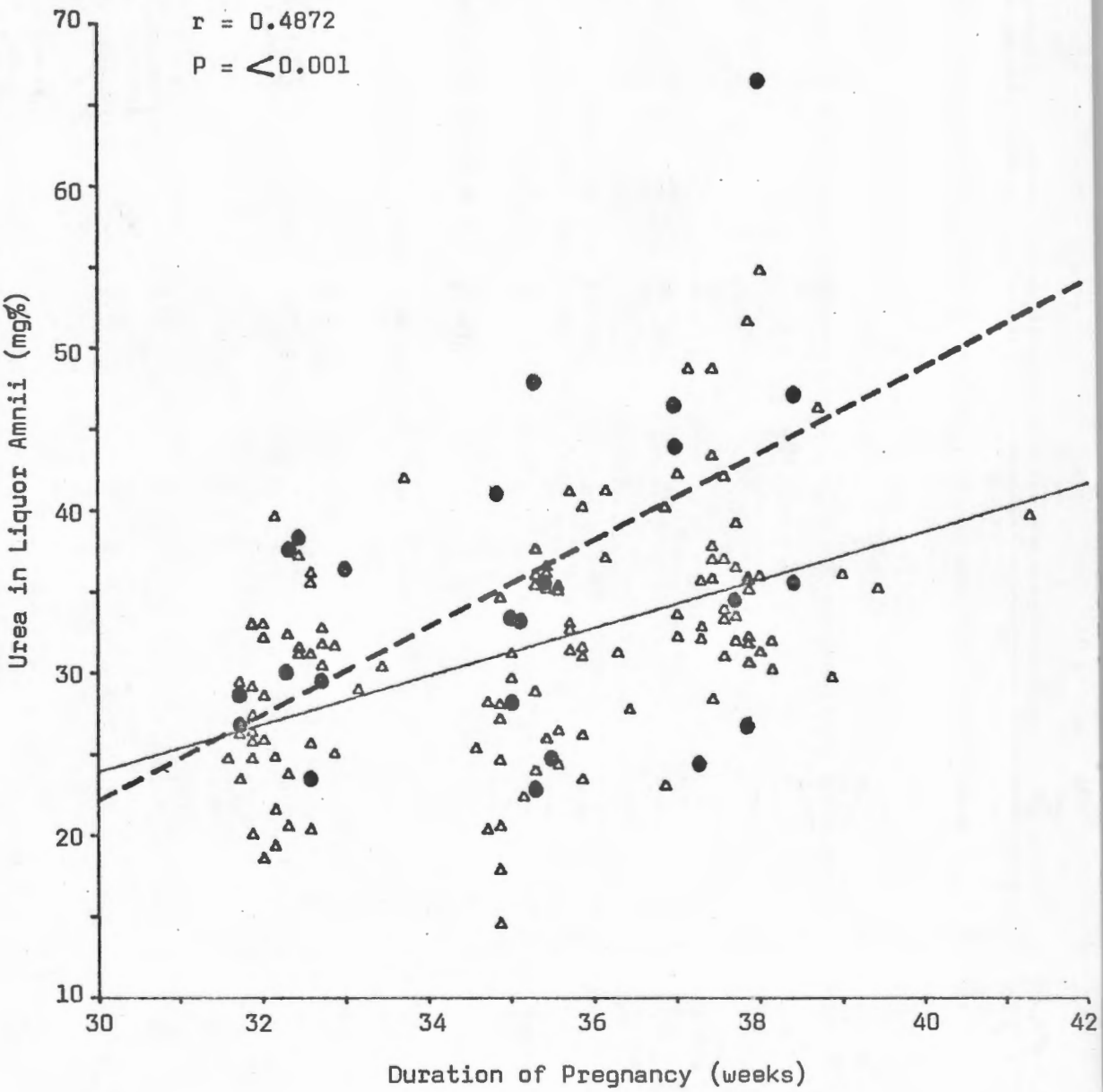
From Figure 38 it would appear that there is no relationship between liquor urea concentration and birth weight.

A marginally better relationship exists between the difference between maternal serum urea concentration and amniotic fluid urea concentration and gestational age, than amniotic fluid urea alone (Figure 39). Segmental analysis of the difference in concentrations reveals only that a difference of 6.5mg% or less indicates a pregnancy of less than 36 weeks.

C. Liquor Creatinine Concentration

Uyeno (1919) concluded that creatinine was absent from amniotic fluid at term and it was not until Pitkin and Zwirek (1967) reported a rise in the concentration of creatinine in liquor amnii with increasing gestation, following the report of McGauchey et al (1960), that this substance was investigated by other workers (Begneaud et al, 1969; Pitkin, 1969; Parmley and

Figure 37



The relationship between the concentration of urea in the liquor amnii and the duration of pregnancy.

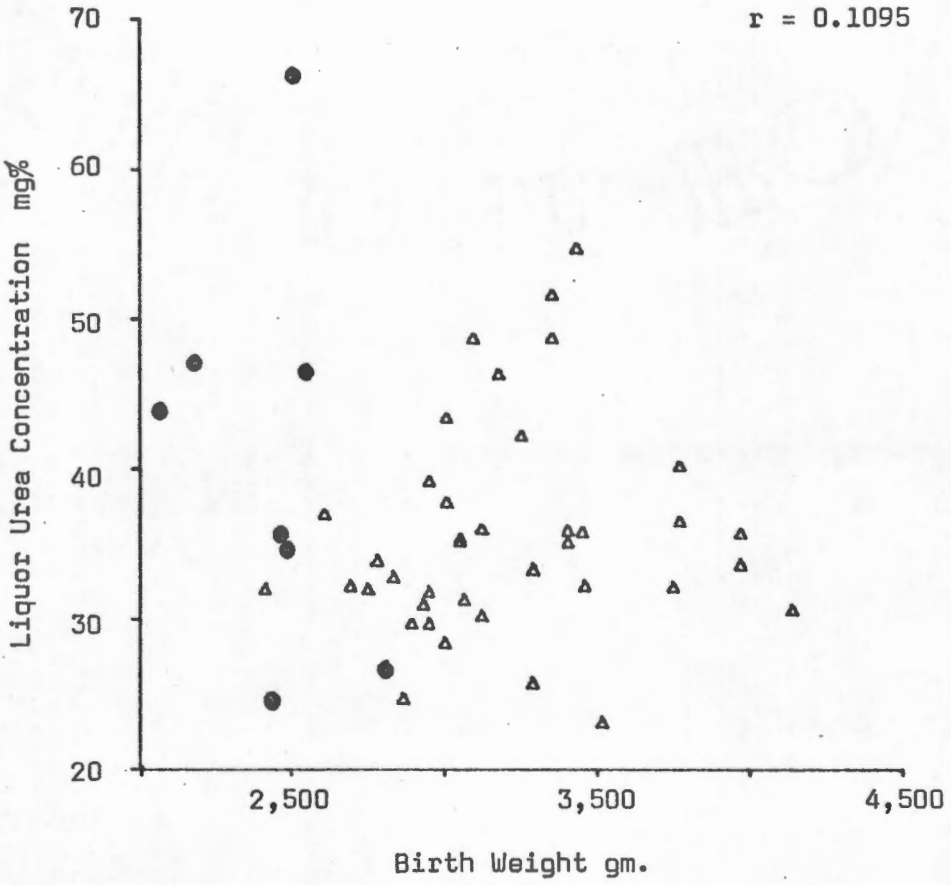
△ Pregnancies resulting in 'normal' infants

● Pregnancies resulting in 'light-for-dates' infants

——— regression line of 'normal' results

– – – – regression line of 'light-for-dates' results

Figure 38

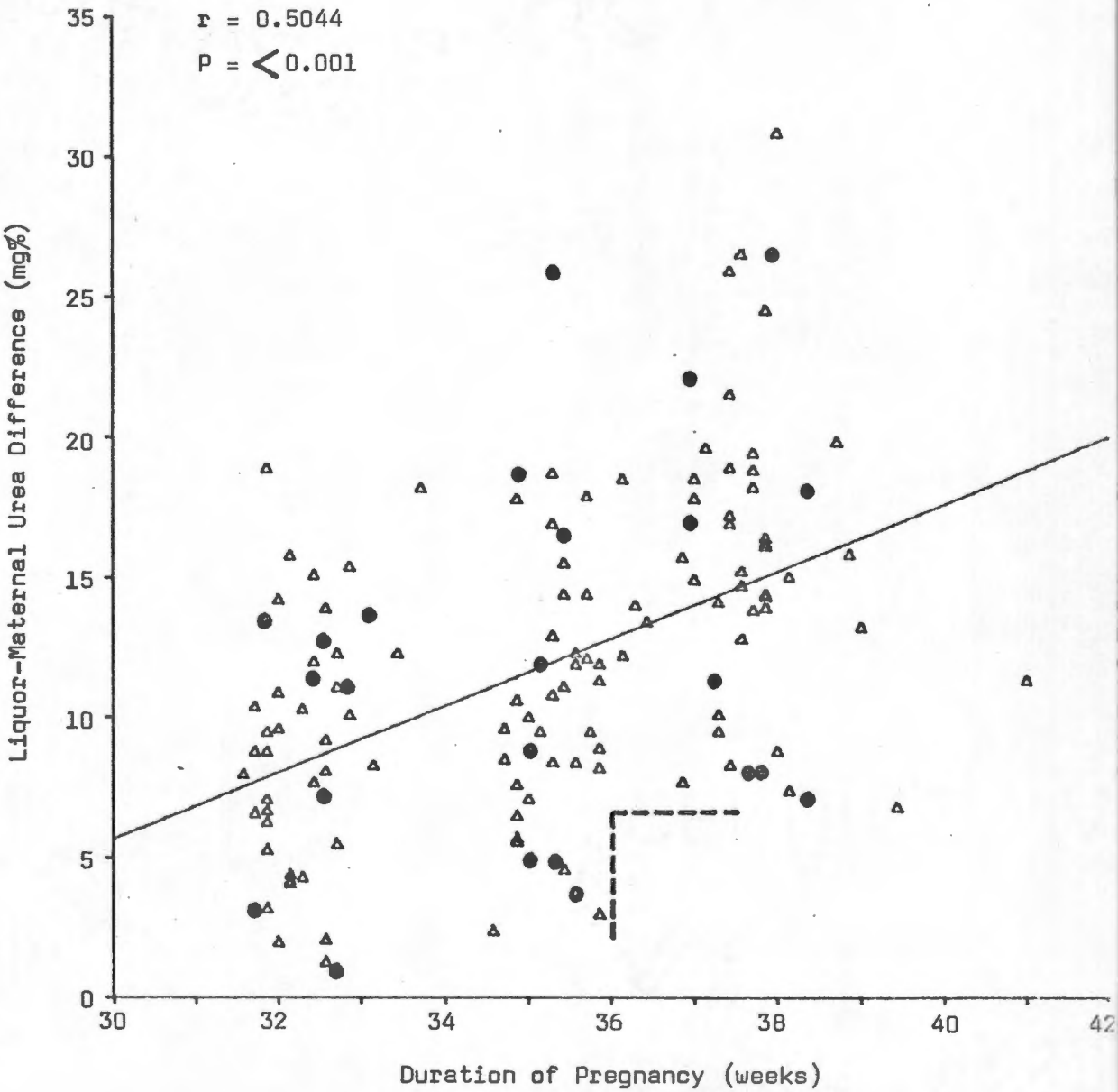


Liquor Urea concentration at the time of the last amniocentesis plotted against birth weight.

△ Normal infants

● Light-for-dates infants

Figure 39



Liquor-maternal urea concentration difference against gestation.

△ Normal infants

● 'Light-for-dates' infants

Miller, 1969; Cherry and Dolger, 1969; Roopnarinesingh, 1970; Doran et al, 1970; Bentram et al, 1970; Henneman et al, 1970; Nelson et al, 1970; Wyatt et al, 1969; White et al, 1969; Droegemueller et al, 1969). Table X lists the findings of these authors.

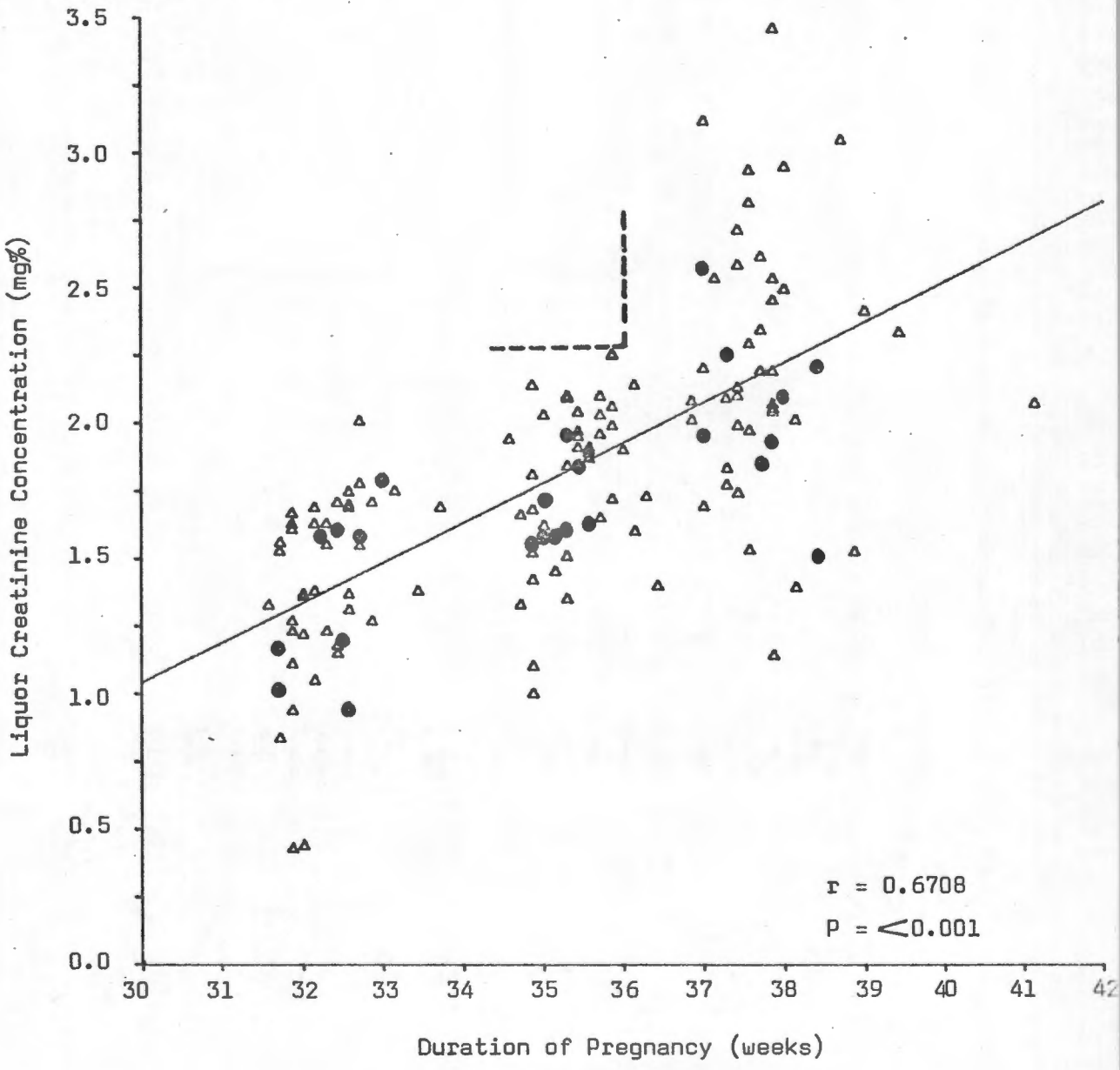
TABLE X

Interpretations attached to specific concentrations of liquor creatinine found in the literature.

Author		Concentration mg%	Interpretation
Bentrem et al	(1970)	>1.5	>36 weeks gestation
Nelson et al	(1970)	>1.5	>36 " "
White et al	(1969)	>1.5	>35 " "
Doran et al	(1970)	>1.6	$\geqslant 37$ " "
Droegemueller et al	(1969)	>1.8	>36 " "
Pitkin and Zwirek	(1967)	>2.0	>37 " "
Begneaud et al	(1969)	$\geqslant 2.0$	$\geqslant 37$ " "
Wyatt et al	(1969)	>1.5	$\geqslant 2{,}500$gm birth weight
Pitkin	(1969)	>2.0	$>2{,}500$gm " "
Parmley and Miller	(1969)	>2.0	$>2{,}500$gm " "

The results of this study confirm the rise in liquor creatinine concentration during the last 10 weeks of pregnancy from a mean of 1.39 ($\pm$ 0.35)mg% at $\pm$ 32 weeks to a mean of 2.23 ($\pm$ 0.5)mg% at 37+ weeks gestation (Figure 40). The 'light-for-dates' pregnancies resulted in a mean liquor creatinine concentration of 1.38 ($\pm$ 0.31)mg% at $\pm$ 32 weeks and 2.03 ($\pm$ 0.30)mg%

Figure 40



Liquor creatinine concentration plotted against the duration of pregnancy. The regression line applies to 'normal' pregnancies only.

△ Pregnancies resulting in 'normal' infants

● Pregnancies resulting in 'light-for-dates' infants

at 37+ weeks. There is no statistically significant difference between these means. A level of 2.25mg% appears to indicate the difference between those above and those below 36 weeks gestation. i.e. no infant had a gestational age of less than 36 weeks when the liquor creatinine concentration was above 2.25mg%. Unfortunately 64% of those infants over 36 weeks gestation had a level of less than 2.25mg%, so that the reverse contention is not valid (Figure 40).

The reason why the results obtained in this study are generally higher than previously reported is perhaps explained by two factors. Firstly, all the pregnancies in this series were normal clinically and virtually every other published series contains an admixture of pathologies. Secondly, the biochemical estimations were done by hand whereas in every reported series the auto-analyzer was used resulting in questionable accuracy but unquestionable reproducibility.

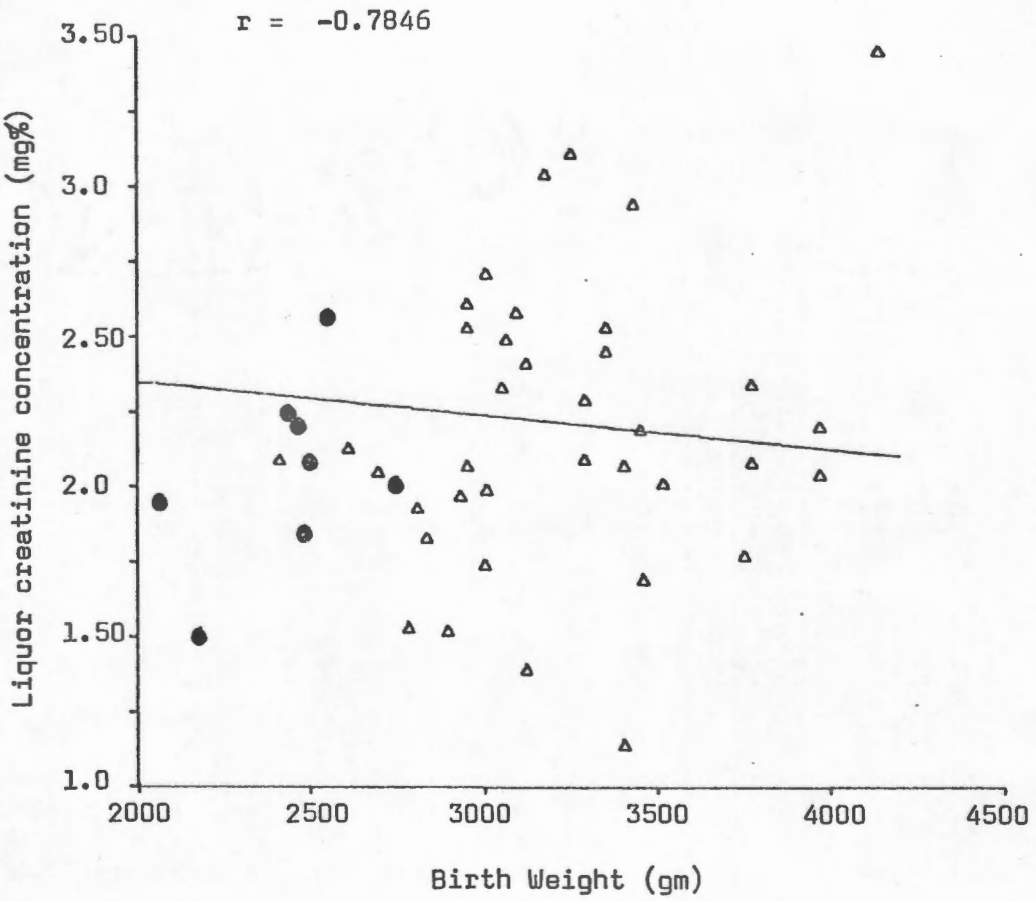
An attempt was made at prediction of birth weight from the last available liquor creatinine value (Figure 41) but the results obtained preclude any such prediction. As an explanation, the author feels that had the biochemical estimations been done within a week of delivery, such a prediction might have been possible. The liquor creatinine values of the three patients who had estimations done upon the day of delivery are seen in Table XI.

TABLE XI

The liquor creatinine concentrations on the day of delivery and the birth weight of the neonate

Case Number	Liquor creatinine concentration (mg%)	Birth Weight (gm)
26	2.32	3050
30	3.03	3175
37	2.06	2950

Figure 41



The relationship between the liquor creatinine concentration at the time of the last amniocentesis and birth weight.

△ Normal infants

● 'Light-for-dates' infants

No conclusions can be drawn from these three results but they suggest a birth weight of over 2,800gm (over 6lbs.) when the concentration of creatinine in the liquor amnii is over 2.00mg%.

D. Liquor True Creatinine Concentration

Because of the author's interest in detailed biochemical changes, a chemically specific technique was used for the estimation of creatinine. This was done in addition to the routine method of estimation and resulted in two separate creatinine results i.e. "true" creatinine and "total" creatinine (Lind, 1969).

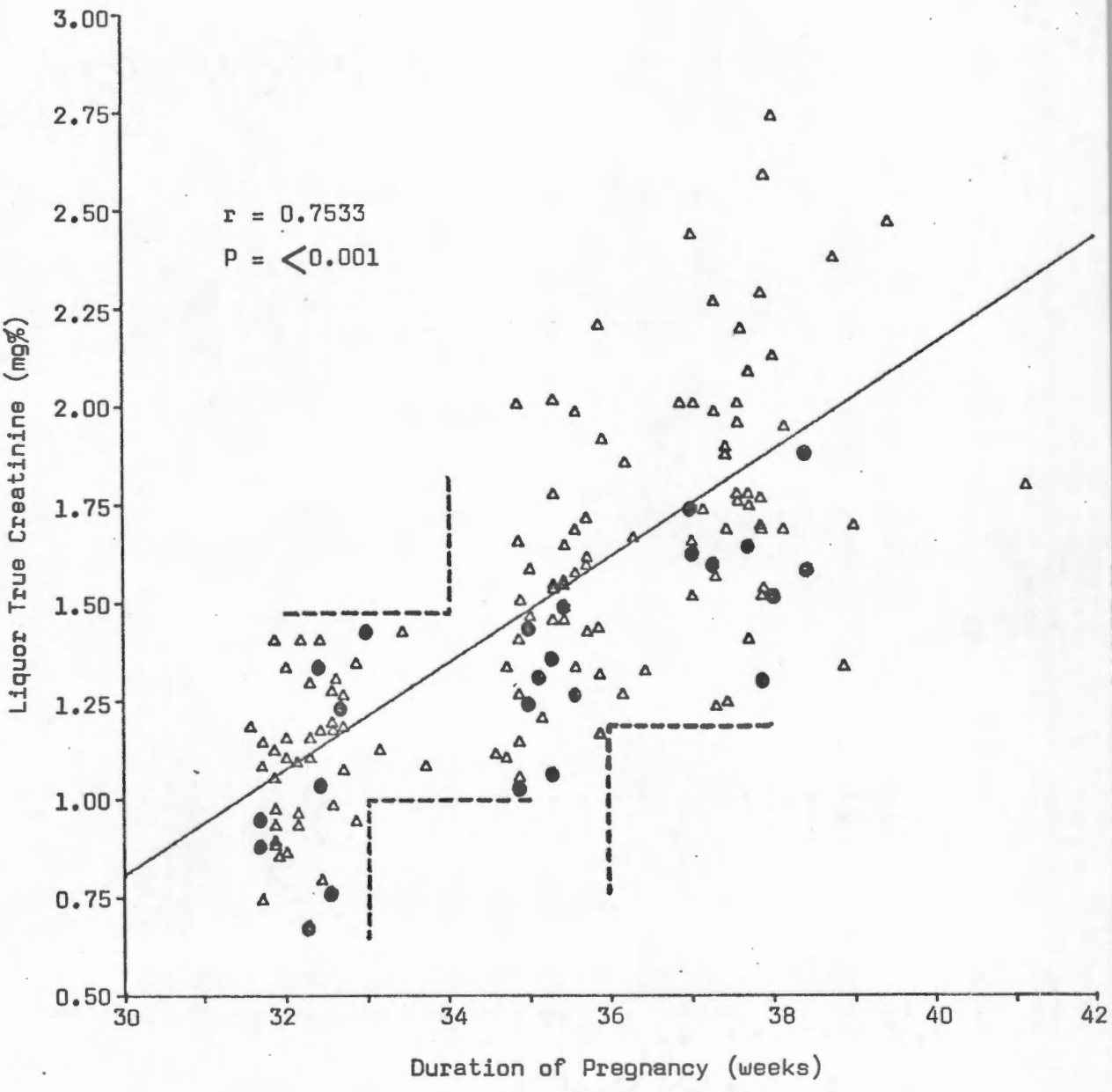
A closer correlation was found to exist between the "true" creatinine concentration in the amniotic fluid and gestational age than with total creatinine i.e. $r = 0.75$ against $r = 0.67$ (Figure 42). No statistically significant difference was found between those pregnancies resulting in normal infants and those resulting in 'light-for-dates' infants. The standard error of the mean was used to compare the mean concentrations of the two groups in each time period.

Segmental analysis of Figure 42 reveals that a concentration of less than 1.00mg% indicates a pregnancy of less than 33 weeks and a value of more than 1.40mg% indicates a gestational age of not less than 34 weeks. All pregnancies of more than 36 weeks gestation had a liquor true creatinine concentration of more than 1.20mg%.

Prediction of birth weight (Figure 43) was again not possible except to say that no fetus of less than 3,000 grams had a liquor true creatinine concentration of more than 2.00mg%.

The creatinine estimation performed upon the maternal serum was the "true" creatinine and thus the difference between the concentrations found in the liquor amnii and the maternal serum

Figure 42

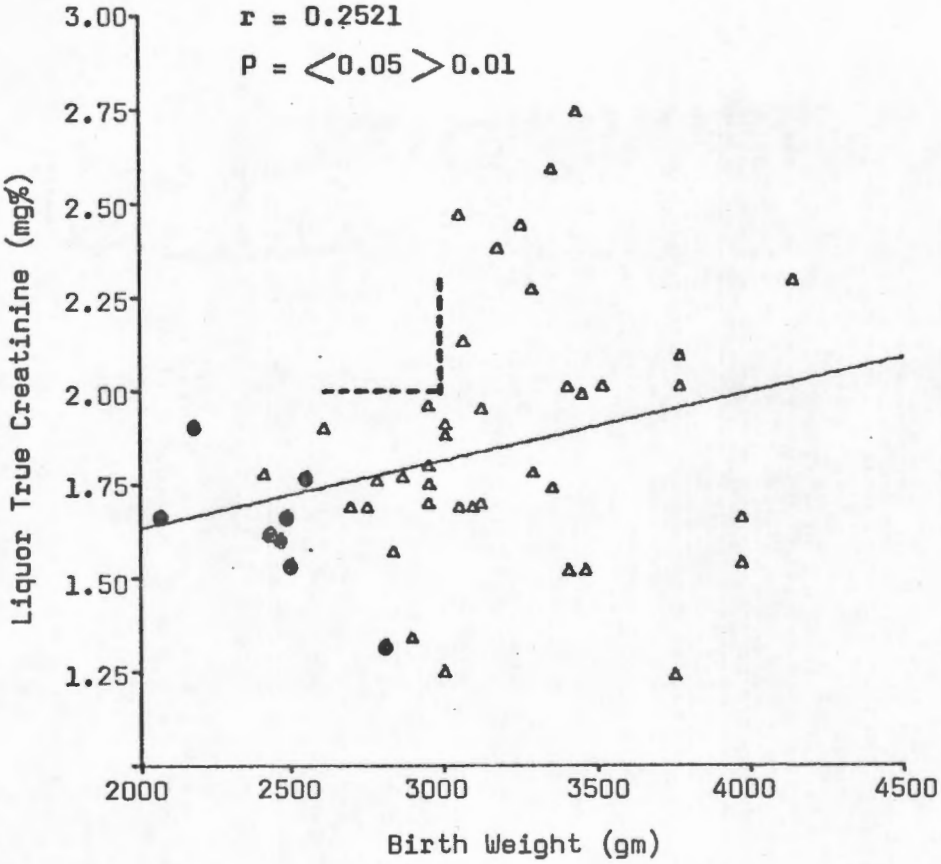


Concentration of true creatinine in the liquor amnii during the last 10 weeks of pregnancy. The regression line applies to 'normal' pregnancies only.

△ Normal infants

● 'Light-for-dates' infants

Figure 43



The relationship between the liquor true creatinine concentration at the time of the last amniocentesis and birth weight.

△ Normal infants

● 'Light-for-dates' infants

could be obtained and analysed. The liquor concentration of true creatinine was never less than that in the maternal serum upon the same day. The difference between the two correlated well with gestational age although the range became larger with advancing gestation (Figure 44). The mean values obtained from the 'light-for-dates' pregnancies did not differ significantly from the means obtained from the normal pregnancies.

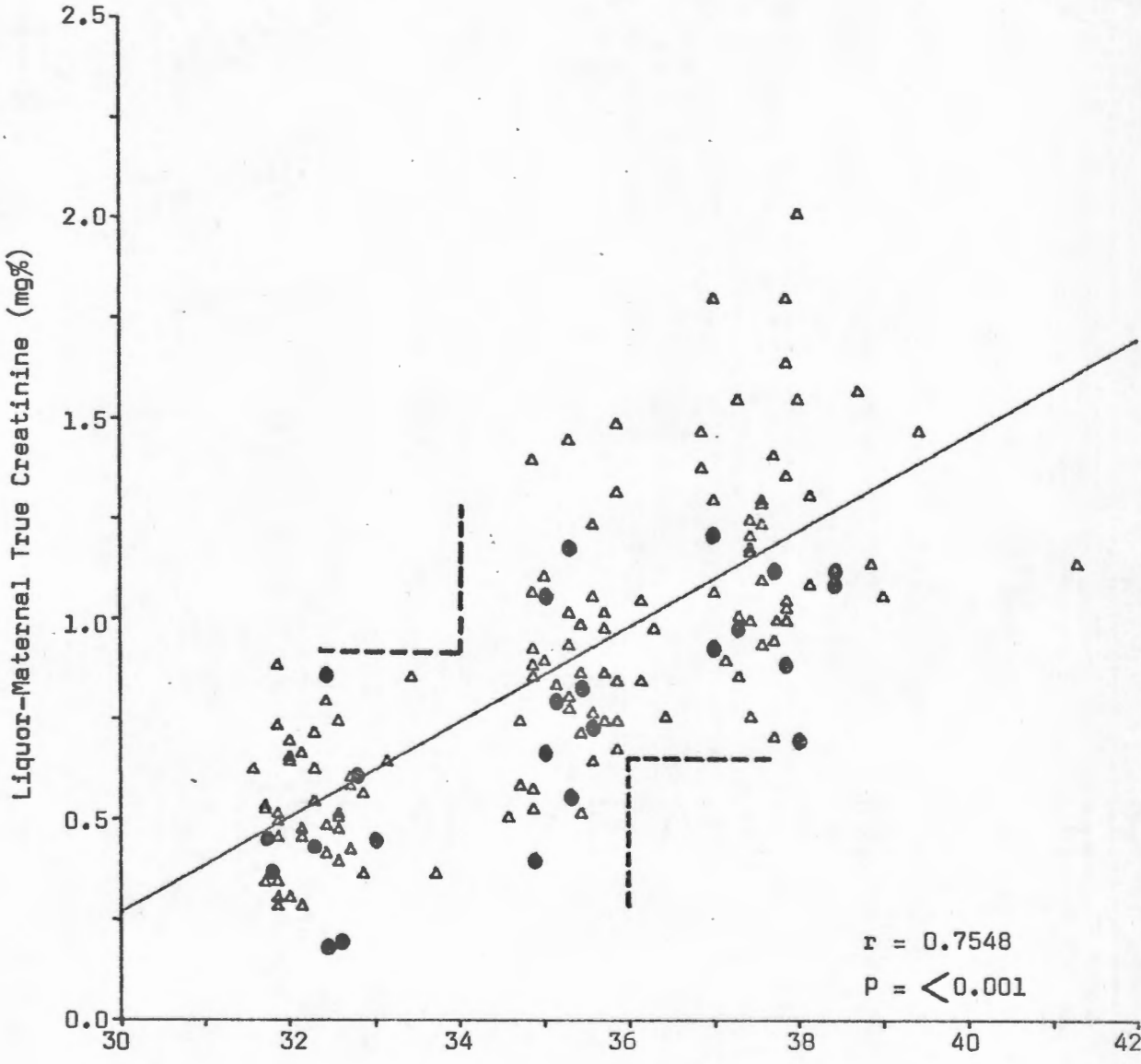
No pregnancy in which the value of the difference in the two estimations was more than 0.9mg% was less than 34 weeks in duration. Similarly no fetus was of more than 36 weeks gestational age when the difference between the two concentrations was less than 0.65mg%.

E. Liquor Bilirubin Concentration

Following the work of Bevis (1950, 1952, 1953 and 1956) the analysis of liquor amnii has come to play a major prognostic role in the management of erythroblastosis fetalis (Fuchs and Cederqvist, 1970). In the antenatal management of this disease, some Rhesus negative patients who were carrying unaffected or Rhesus negative infants had amniocenteses and some workers reported very low spectrophotometric readings in these cases (Walker, 1957; Walker and Jennison, 1962; Goldstein et al, 1966). Mandelbaum et al (1967) reported that the 450mμ peak in the absence of erythroblastosis fetalis disappeared after 36 weeks of gestation. They found a good correlation with gestational age and 85% of their cases with a spectrophotometric reading of zero produced babies or more than 6lbs. They suggested that this finding was a function of fetal hepatic enzyme maturity.

Parmley and Miller (1969), Bentrem et al (1970) and Henneman et al (1970) have all found a decrease in the spectro-

Figure 44



Liquor-maternal true creatinine difference against gestation

● 'Light-for-dates' infants

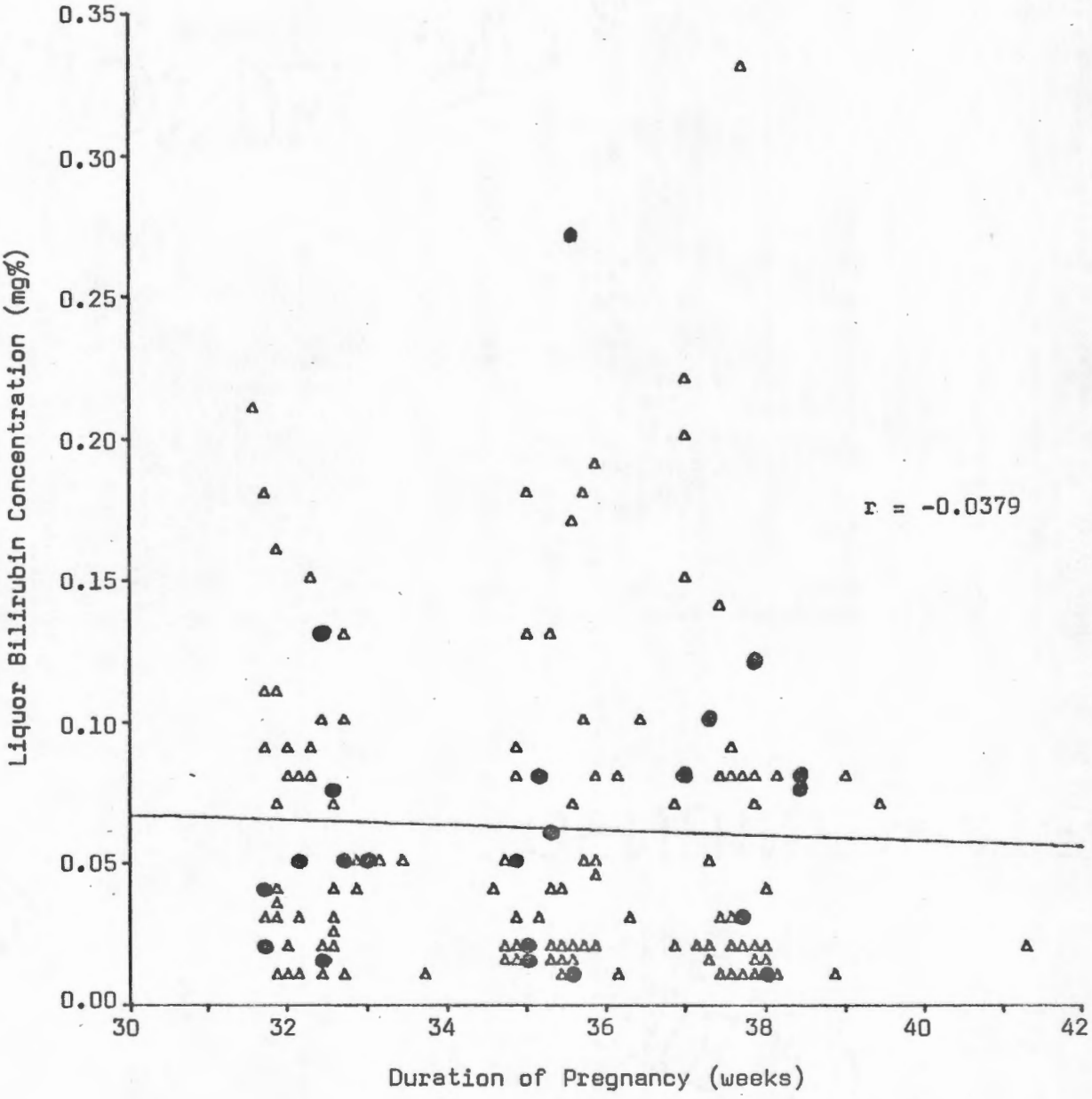
△ Normal infants

photometric peak at 450mμ with advancing gestation but claim that the results are variable and that this estimation is of no value in the assessment of gestational age.

Oxyhaemoglobin, meconium and methaemalbumin have all been found to produce interference at very similar wavelengths to bilirubin when the liquor amnii is examined spectrophotometrically (Clayton et al, 1969; Black et al, 1969; Halitsky and Krumholz, 1970). For this reason it was decided to estimate the concentration of bilirubin biochemically although the method had an experimental error of 12% (Appendix C).

The results of these estimations have been plotted against gestational age in Figure 45. The regression line indicates a very gradual decline but the range of values at any given week of gestation precludes the use of this parameter in the estimation of gestational age. This does not necessarily mean that the concentration of bilirubin in the amniotic fluid is not an index of something other than gestational age or birth weight, or even that spectrophotometric analysis for bilirubin does not provide an index of gestational age. From the results in this study, however, no such information can be gained from the concentration of bilirubin in the liquor amnii.

Figure 45



Concentration of bilirubin in the liquor amnii during the last 10 weeks of pregnancy.

△ Normal infants

● 'Light-for-dates' infants

8. THE COMPLICATIONS ASSOCIATED WITH AMNIOCENTESIS

Because the amniocenteses in this project were done for purely research purposes, it was incumbent upon the author to eliminate all possible sources of risk from the procedure. The British Medical Research Council has recommended that amniocentesis should not be undertaken on normal pregnant women for research purposes only (British Medical Journal 1971: 4, 245). The author had already concluded his study when this recommendation was published, but feels that because the analysis of liquor amnii appears to yield valuable parameters of gestational age, all the complications encountered in this study should be evaluated.

A. Aspiration of blood or blood-stained liquor

This complication of amniocentesis would appear to be one the commonest reported in the medical literature although agreement as to its incidence is conspicuously absent. Walker and Jennison (1962) reported an incidence of 5% in 500 amniocenteses, whilst Queenan and Adams (1964) had an incidence of 66%. Possible factors concerned in the variation in incidence of this complication are: experience in the procedure, the actual technique used and even the definition of blood-staining of liquor used by the investigators.

The author was believed by the senior members of his department to have acquired sufficient experience in the technique during his training and it was felt that the technique to be used (Chapter 3, section 7, page 15) was the safest possible. The definition of blood-staining of the liquor was divided into two parts viz. (i) overt macroscopic blood-staining and (ii) 'Microscopic' blood-staining i.e. the discovery in liquor, initially

thought to be free of blood, of a minute quantity of blood at the bottom of the pellet of cells and debris obtained by centrifuging the specimen.

In view of the knowledge that deterioration of haemolytic disease as a consequence of obtaining blood-stained liquor can occur (Queenan and Adams, 1964; Grobbelaar and Trott, 1968) it was felt that knowledge of the incidence of all blood staining was important and thus the 'microscopic' category was introduced (Bennett, 1972b). The aspiration of pure blood signifies that the needle is either within the substance of the myometrium or the placenta.

In 139 amniocenteses performed on the 47 patients in this study, 31 samples of blood-stained liquor amnii were obtained, an incidence of 22.3%. Of these, 15 were macroscopically contaminated with blood (10.7%), and the other 16 (11.6%) were 'microscopically' stained. By means of the acid elution technique (Chapter 3, section 11, page 34), it was possible to subdivide the type of blood so obtained into four groups.

TABLE XII

A breakdown of the type and frequency of blood-staining of the liquor resulting from 139 amniocenteses.

Type of Blood Staining		Type of Blood			
		M	F	M>F	F>M
Macroscopic	15	5	4	2	4
Microscopic	16	12	0	3	1

M : Maternal blood only F : Pure fetal blood

M>F : An admixture in which maternal blood predominated

F>M : An admixture in which fetal blood predominated

With the passage of time it became obvious that experience in the technique was of obvious importance with particular reference to this complication. This is illustrated by the fact that in the first 54 amniocenteses in the series, seven were complicated by macroscopic and 11 by microscopic blood-stained amniotic fluid. In the remaining 85 amniocenteses, the figures were eight and five respectively.

B. Feto-Maternal Haemorrhage

As has already been described, samples of maternal blood were obtained before and after each amniocentesis for study by the acid elution technique (Chapter 3, section 11, page 34). An interesting finding was that five (10.6%) of the 47 patients had demonstrable fetal cells in their circulations prior to the first amniocentesis at ± 32 weeks.

Of the 139 smears taken 'post' amniocentesis, 14 (10.1%) were found to contain fetal cells. From these figures it can be concluded that amniocentesis increases the natural incidence of feto-maternal micro-transfusion by ± 100%.

For the purposes of this study, a count of 15 or more fetal cells per 35 low-power fields was accepted as representing 1.0ml or more of fetal blood in the maternal circulation (Chapter 3, section 11, page 34). This was defined as a major transplacental haemorrhage. Of the 14 positive acid elution smears, only two were found to have more than 15 cells and thus constitute a major feto-maternal bleed by the above definition. These two smears had fetal cell counts of 17 and 41 cells each, corresponding to 1.25ml and 3.0ml of fetal blood in the maternal circulation respectively.

When considering the association between blood-staining of

the liquor of both types and the incidence of transplacental haemorrhage, it was found that a bloody tap had been obtained in six of the 14 feto-maternal haemorrhages in contradistinction to the report of Woo Wang et al (1967). Five of these were associated with blood-staining of the microscopic type. From Table XII it can be seen that in only four of the 'microscopic' blood-stained specimens were fetal cells present. The conclusion to be drawn is that either the arbitrary differentiation between the two types of blood-staining is not a practical one, or, that in the one case, fetal cells escaped into the maternal circulation but not into the liquor amnii.

C. Accidental Haemorrhage

This complication is uncommonly reported (Grobbelaar and Trott, 1968). In this series of patients, one patient (2.1%) presented with an abruptio placentae following an amniocentesis. This occurred during the 37th week of pregnancy when an external cephalic version was unwisely followed within 30 minutes by an amniocentesis. Mild abdominal pain and vaginal bleeding occurred 12 hours later and an immediate Caesarean section averted what might have easily been a tragedy.

Although external cephalic version is a well recognised, though uncommon, cause of abruptio placentae (Donald, 1969), amniocentesis in the above circumstances must shoulder at least half the blame. A combination of these two procedures upon the same patient on the same day is a mistake the author is unlikely to ever repeat.

D. Infection

Various authors (Liley, 1960; Alpern, Charles and

Friedman, 1966; Fairweather and Walker, 1964; Walker, 1970) have reported the occurrence of both chorio-amniitis and the unexplained feature of maternal pyrexia and rigors. The author believes the absence of these complications from this series is due to the rigid adherence of sterile precautions.

E. Failure

This occurred once in this series and was due to faulty technique and excessive haste on the part of the author. The careful selection of the site of puncture reduces this problem to a minimum but it does nevertheless occasionally happen (Gordon, 1969). One patient went into spontaneous labour and delivered one day prior to the date upon which her last amniocentesis was due.

F. Premature Labour

Before the start of this study, it was decided that should any labour occur within 48 hours of an amniocentesis, then the procedure would be regarded as the aetiological factor.

Fortunately no patient in this study group went into labour within 48 hours of amniocentesis.

Based upon the above definition, amniocentesis would seem to be a rare cause of the onset of premature labour. The author feels that the onset of labour within six days of amniocentesis as reported by Liley (1960) or within five days (Alpern et al, 1966), does not take into account those labours which might have started spontaneously within those time periods and which, therefore, are unrelated to amniocentesis.

9. RADIOLOGICAL ASSESSMENT OF GESTATIONAL AGE

Several radiological techniques have been advocated to assess fetal development in an attempt at determining gestational age. Initial attempts at predicting fetal weight, when the latter was used as an index of gestational age, were unsuccessful (Donaldson and Cheney, 1948). Morley (1961) concluded that accurate prediction of fetal weight could not be achieved by any radiological method.

Christie (1949) X-rayed infants shortly after birth and noted the effect of race and sex upon their osseous development. In a follow-up study (Christie et al, 1950) a correlation was found between the appearance of the epiphyses of the knee joint and the duration of pregnancy. Adams (1955) found that the lowest perinatal mortality correlated well with the presence of a radiologically demonstrable distal femoral ossification centre. Moir (1955) suggested that specific ossification centres appeared at particular times during pregnancy but stressed that these times were at best only averages and liable to much individual variation.

Hartley (1957) laid down criteria for the radiological estimation of fetal maturity claiming that certain ossification centres developed at specific stages in pregnancy. These criteria have been widely accepted (Willocks, 1971). In spite of the specificity of the time of development of these centres claimed by Hartley (1957), various authors reported a wide range of results using this method (Murdoch and Cope, 1957; Cope and Murdoch, 1958; Berridge and Eton, 1958; Schreiber et al, 1962; Bluth, 1966; Dee et al, 1966).

Various factors have recently been claimed to affect the osseous development of the fetus by either advancing or retarding it. Scott and Usher (1964) demonstrated the retardation of ossification

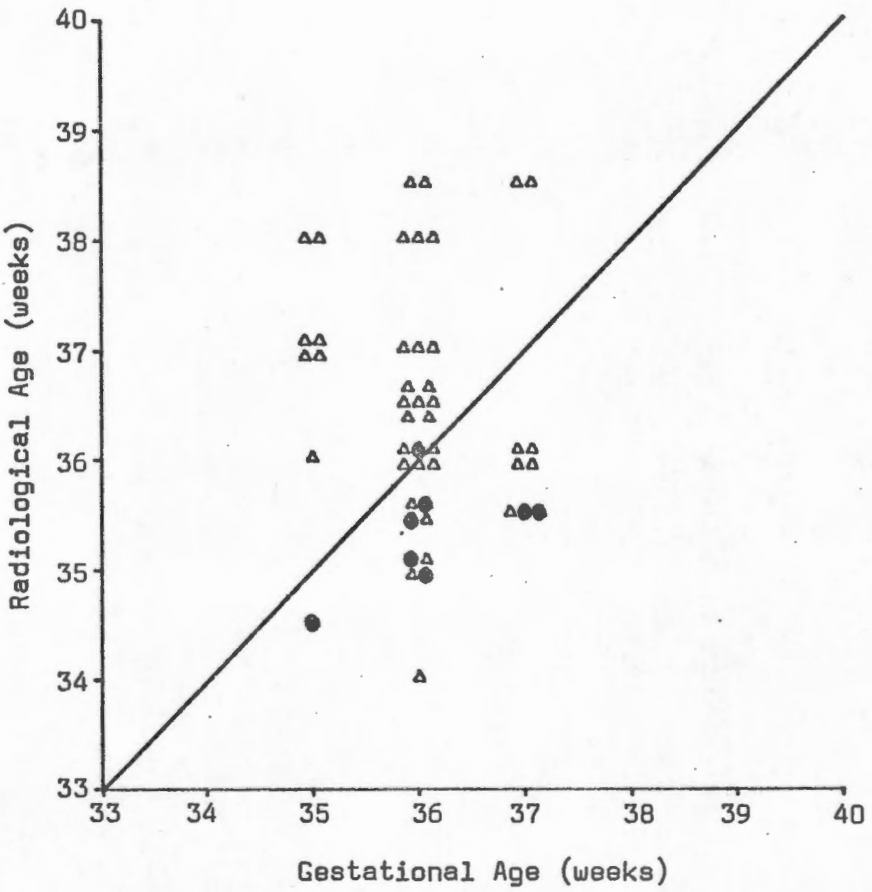
of the epiphyses of the knee joint in infants subjected to 'intra-uterine malnutrition'. Usher (1966) found that 80% of malnourished infants had no radiologically demonstrable proximal tibial epiphysis at or after 38 weeks. On the other hand Russell and Rangecroft (1969) maintain that the rate of epiphyseal maturation is undisturbed by factors causing low birth weight and that dysmature babies cannot be diagnosed antenatally by delay of maturation. Russell (1969) was unable to confirm the earlier findings of Christie (1949) and found that maternal parity, maternal age, sex of infant and socio-economic status of the parents did not affect the osseous development of the infant. The only factors able to influence this development that Russell and Rangecroft (1969) were able to confirm were the delayed development associated with spina bifida of the fetus, and the advanced development in infants of insulin-dependent diabetic mothers and in association with anencephaly. More recent studies, however, (Usher, 1970; Croall and Grech, 1970) appear to confirm the earlier work of Scott and Usher (1964) and Usher (1966) in that factors leading to 'light-for-dates' or dysmature infants do, in fact, lead to retarded bone ossification. Russell and Rangecroft's study was a computerized one based entirely upon the menstrual histories supplied by the patients, and is, therefore, open to criticism.

All the X-rays in this study were assessed by a Senior Consultant in the Department of Radiology. As all X-rays were taken at 36 ($\pm$ 1) weeks of gestation, radiological ageing was assessed upon the presence or absence of the epiphyses round the knee joints (Hartley, 1957). The results of this assessment are diagramatically represented in Figure 46.

Allowing the radiologist $\pm$ 1 week error, it was found that 27.6% of fetuses (13) were radiologically over-assessed and 8.5% (4) were under-assessed. Two of the four were 'light-for-dates' infants.

The diagonal line through the points on the graph represents

Figure 46



Comparison of radiological assessment of gestational age and gestational age in completed weeks calculated from the L.N.M.P.

△ Normal infants

● 'Light-for-dates' infants

the line along which all the assessments should lie were there perfect correlation. It will be seen that no 'light-for-dates' fetus was ascribed a radiological age of more than its calculated age. Remembering that the criteria for the diagnosis of 'light-for-dates' infants was a very rigid - 1 standard deviation from the mean weight for age, these results tend to support the contention of Scott and Usher (1964) that the factors responsible for the 'light-for-dates' infant produce delayed ossification of epiphyses.

This very small study does not support the theory of Russell (1969) that the radiological appearance of the distal femoral epiphysis should be regarded as occurring at 37 weeks and not 36 weeks.

10. DISCRIMINANT ANALYSIS

No single parameter has yet been found reliable as an index of gestational age for the following reasons:-

i. many patients are unreliable at remembering the date of their last menstrual period.

ii. clinical parameters have been shown to be difficult to quantitate and liable to much variation, either subjective or because of some pathological state of pregnancy.

iii. special investigations show wide variations and yield both false positives and false negatives. This is because of differences in methodology and interpretation of results, as well as uncertain effects of different pathological states of pregnancy upon the parameters measured. The most difficult aspect of special investigations would seem to be the interpretation of the large biological variations encountered.

Many authors have, therefore, attempted various combinations of parameters in an effort to arrive at a more accurate and clinically useful answer (Brosens et al, 1969; Murphy, 1969; Pitkin, 1969; Mandelbaum and Evans, 1969; Wyatt et al, 1969; Horger and Hutchinson, 1969; Parmley and Miller, 1969; Bentrem et al, 1970; Henneman et al, 1970; Nelson et al, 1970; Turnbull, 1970; Lind, 1970 (personal communication); Casaer and Akiyama, 1970).

From the preceeding results, no single parameter was considered a reliable index of gestational age, even in the group of normal pregnancies studied. For this reason, and because it was felt that different parameters might be applicable to different periods of

gestation during the last 10 weeks thereof, it was decided to subject the results obtained in this study to stepwise discriminant analysis. The statistical problem posed by the dependence of the parameters upon each other by virtue of their serial measurement in the same patients has already been discussed.

A. Preparation of Data

1. Firstly three sets of observations on each of the 39 normal patients were used to provide 115 observations (variables), 39 in each of the first two time groups, and 37 in the last. The variable which, when used alone, best enables discrimination between the time groups is then selected i.e. that variable which minimises the ratio of variance within groups to total variance. Thereafter variables are added in successive steps until further addition of variables does not improve the discriminant function. At each step the discriminant function is used to classify all the cases given, giving an idea of how effective it is.

2. To eliminate the effect of dependence, the cases (patients) were divided up randomly into three groups and only one set of variables taken from the same patient in each time interval. The analysis was then repeated on these smaller samples. This was repeated twice.

3. Friedman's two-way analysis of variance was applied to each variable in an attempt to identify those variables with a time defect. All variables were subsequently subjected to stepwise discriminant analysis resulting in the results of the Friedman test being discarded.

B. Results

Table XIII shows the variables in the order in which they were added until no further addition made a significant difference at the 5% level to the discrimination.

TABLE XIII

Variables in the order in which they were added to the discriminant function and percentage correct classification at each step

Step Number	Variable entered	Percentage correct classification of cases
1	Ultrasonic Biparietal Diameter	74%
2	Clumping of fat cells	80%
3	Difference between liquor and maternal true creatinine concentration	84%
4	State of Cervix	87%
5	Difference between liquor and maternal urea concentration	87%
6	Liquor urea concentration	88%

The three discriminant functions for each time interval are:

$$X_1 = 9.95812y_1 - 0.91317y_2 - 6.42893y_3 + 10.90166y_4 + 1.20859y_5 - 0.82589y_6 - 433.25195$$

$$X_2 = 10.66992y_1 - 1.29052y_2 - 0.25351y_3 + 11.49984y_4 + 1.46688y_5 - 1.05281y_6 - 498.62988$$

$$X_3 = 11.02552y_1 - 3.16949y_2 + 1.70368y_3 + 13.65465y_4 + 1.68888y_5 - 1.05137y_6 - 542.68848$$

Where X_j denotes the discriminant function of the j^{th} time interval and y_i denotes the value of the variable entered at the i^{th} step.

When considering the effect of dependence of the variables upon each other, two subsamples were subjected to the same analysis. The results of these two analyses are shown in Tables XIV and XV.

TABLE XIV

Variables in the order in which they were added to the discriminant function

Step Number	Variable entered	Percentage correct classification of cases
1	Ultrasonic Biparietal Diameter	68%
2	Clumping of fat cells	84%
3	Uterine volume	84%
4	Liquor creatinine concentration	89%
5	Liquor bilirubin concentration	92%

Using the same notation as before the discriminant functions are:

$$X_1 = 12.35796y_1 - 14.48029y_2 + 0.01439y_3 - 5.58939y_4 + 194.73596y_5 - 571.34888$$

$$X_2 = 13.12753y_1 - 16.24771y_2 + 0.01604y_3 - 2.33281y_4 + 211.61687y_5 - 653.90674$$

$$X_3 = 13.37006y_1 - 6.85109y_2 + 0.01927y_3 + 4.63542y_4 + 182.16185y_5 - 711.11011$$

TABLE XV

Variables in the order in which they were added to the discriminant function

Step Number	Variable entered	Percentage correct classification of cases
1	Uterine volume	76%
2	Clumping of fat cells	76%
3	Difference between liquor & maternal true creatinine	89%

Using previous notation the discriminant functions are:

$$X_1 = 0.00680y_1 + 0.39370y_2 + 6.60065y_3 - 15.59532$$

$$X_2 = 0.00873y_1 + 0.94491y_2 + 16.10884y_3 - 30.96155$$

$$X_3 = 0.01073y_1 + 7.63042y_2 + 17.52272y_3 - 47.13383$$

It is important to realise that the variables included in a discriminant function are not necessarily all those which vary most between time intervals. If two variables are highly correlated and both able to indicate the time interval, when one is selected the other may have little to add to the discrimination. The discriminant functions should, therefore, be treated as a method of classification and not as a list of variables which are related to the age of the fetus.

All three of the above analyses predict correct classification of the pregnancies studied into one of the three time intervals with 89.7 ($\pm$ 2.1)% accuracy. Because the factor of dependence tends to invalidate the results given in Table XIII, although the sample is very much larger than the two sub-samples,

it was decided to test further the results given in Tables XIV and XV.

The two sets of discriminant functions were obtained using a stepwise procedure on a sample of the total data, in which observations taken on a patient at only one of the time intervals was used. The data not used in this analysis was next classified using the two sets of discriminant functions in Tables XIV and XV in order to decide which of the two sets was the best.

Each of the three time intervals (30 - 33 weeks, 34 - 36 weeks, and 37+ weeks, called Groups 1, 2 and 3 respectively) was treated separately. The classifications obtained appear in Tables XVI and XVIII. The 'light-for-dates' cases which were originally excluded from the analysis, were also included.

TABLE XVI

Classification of cases according to the discriminant functions of Table XIV

Correct Group	Number of cases classified	Numbers classified into each group		
		1	2	3
1	34	28	6	0
2	34	9	24	1
3	32	0	12	20

TABLE XVII

Classification of cases according to the discriminant functions of Table XV

Correct Group	Number of cases classified	Numbers classified into each group		
		1	2	3
1	34	28	5	1
2	34	10	23	1
3	32	1	16	15

From these tables it can be seen that for classification between groups 1 and 2, there is no significant difference between the discriminant functions of Tables XIV and XV. However, the set given in Table XIV is better for discriminating between groups 2 and 3.

A stepwise discriminant analysis was performed separately on the normal and 'light-for-dates' cases at 30 - 33 weeks, 34 - 36 weeks and 37+ weeks in order to find those parameters which best differentiated between the two groups of patients. The results of this analysis appear in Table XVIII.

TABLE XVII

Results of the Stepwise Discriminant Analysis for the Normal and 'Light-for-dates' cases done separately at 30 - 33 weeks, 34 - 36 weeks and 37+ weeks.

Step Number	30 - 33 weeks		34 - 36 weeks		37+ weeks	
	Variable	% correct classification	Variable	% correct classification	Variable	% correct classification
1	Ultrasonic biparietal diameter	74	Ultrasonic biparietal diameter	85	Uterine volume	81
2			Uterine volume	87	Ultrasonic biparietal diameter	92

(Variables in the order in which they were added until no further addition made the subset significantly better at the 5% level of significance).

CHAPTER 6

DISCUSSION

1. INTRODUCTION

"A woman with suppression of the catamenia may not be pregnant, but no woman who has a foetus in utero can menstruate", said James Hamilton in 1826.

This discussion is intended to cover briefly, the problems faced by the clinical obstetrician in Cape Town at present (1970/71) and, therefore, the reasons for this study, the methods used and any suggestions the author may have for improvements, as well as a critical appraisal of the results, their meaning and application to the field of modern obstetrics.

In spite of Hamilton's categorical statement, it is now appreciated that a pregnant patient can, in fact, menstruate, although the flow and duration of bleeding are often less than normal (Greenhill, 1965; McClure Browne and Dixon, 1970). Donald (1968) claims that up to 50% of women are not absolutely sure of the date of onset of the last menstrual period. Lind (1970) is more generous with a claim that up to 20% of women fall into this category. The author investigated this problem in Cape Town, by checking the calculated gestational age with the paediatric assessment of the age of the neonate, over a two month period at two of the five hospitals in the Peninsula Maternity Services group. The detailed results of this investigation are to be found in the Appendix. In summary, 32.7% of patients who were able to give the date of their last menstrual period were considered to be wrong, whilst 5.1% of the European patients and 17.1% of the non-European patients give no date at all. It would thus appear that between 66.3% and 53.7% of patients in Cape Town are able to provide

the clinician with a reliable index of the duration of their pregnancies. Elective termination of pregnancy accounts for over 14% of our 14,500 annual deliveries. The problem of proper timing of delivery in order to minimise the risks of prematurity is, therefore, a very large one, and one which needed a practical solution. That 15% of the live born infants premature by weight delivered in 1970 were of pregnancies iatrogenically terminated, is a very disturbing fact.

The voluminous literature on the subject of parameters of gestational age which has accumulated over the last decade, indicated to the author that no controlled series of normal patients had been studied in serial fashion. Little, if anything, was known about the normal distribution of any of the parameters reported upon. Examination of the facilities available and the recent acquisition of a diasonograph prompted the author to embark upon such a study.

2. MATERIALS AND METHODS

Retrospectively, the only criticism the author has of the materials used, is that the number of patients was very small. It must be appreciated, however, that the single most difficult task was to persuade normal healthy primigravidae to volunteer to be 'guinea pigs' in a research project from which they could expect to derive no benefit. That neither amniocentesis nor X-ray examination of the fetus could be guaranteed to be harmless procedures and the fact that they were purely for research purposes was stressed to each patient.

It was realised too late in the project that better use should have been made of the existing facilities at the time the study was started. A special laboratory was established for the biochemical estimations and no thought was given to running duplicate samples through the routine service supplied by the Department of Chemical

Pathology. Although this would almost certainly not have influenced the results, it would have provided an excellent guide for the future. At present a Gestational Ageing Service is being provided for the Department of Obstetrics and Gynaecology, and since the closure of the author's laboratory all chemical estimations are being done by the Department of Chemical Pathology. It would be most useful to know how their results would have correlated with those obtained by the author in respect of normal pregnancies.

With regard to the bilirubin concentration estimations, spectrophotometric analysis of the liquor amnii should also have been done simultaneously. Had a correlation between the two methods been found, as surely it must, the time consuming biochemical method could have been advantageously abandoned.

The measurement of liquor volume by means of a dye dilution technique necessitates a prolonged exposure of the intra-amniotic contents to the external environment. The author did not feel that this additional risk was one he could justifiably expose his patients to.

Retrospectively however, the author would have liked very much to have known the volume of the liquor amnii at the time when biochemical investigations were performed. By what means liquor volume is controlled, if it is controlled at all, is unknown but individual variability of volume in late pregnancy is considerable. It, therefore, seems logical that the total amount of urea or creatinine, for example might be more meaningful than their concentration.

3. MOTHERS AND INFANTS

The selection of the patients was made with the express purposes of (i) trying to equalise the socio-economic standards in the two race groups to try as far as practically possible to reduce

the effects of socio-economic factors upon the pregnancies to as low as possible.

(ii) studying a group of normal, healthy primigravida who, at the time of incorporation into the study group, manifested no evidence of pathological deviation from normal pregnancies.

The results in Table III suggest that whatever socio-economic factors were active, their results were very similar in the two racial groups. There was no statistically significant difference between the means of the two racial groups given in Table III.

The criterion by which infants were classified as 'light-for-dates' was a very rigid 1 standard deviation below the mean birth weight for gestational age. This was selected in order that those infants judged as normal weight for age would be accepted as normal by any weight for age standards (Lubchenco et al, 1963; Gruenwald, 1966; Neligan, 1965). The eight infants who were designated as 'light-for-dates' may well have been within the normal distribution for the population from which the sample was taken but the author wanted to be sure he was dealing with normal pregnancies and babies.

It is interesting to note that four non-European infants and four European infants were classified as 'light-for-dates'. At the time of writing it is not known whether this even distribution of 'light-for-dates' infants is representative of the unit as a whole or whether one or other ethnic group tends to have a higher incidence.

Although the complications of amniocentesis have already been discussed (Chapter 5, section 8) it is relevant to discuss the two deliveries alluded to in Table IV.

Patient number: 7 L.N.M.P. 26.3.1970

E.D.D. 2.5.1971

Weight gain of 6.6kg since the time of booking at 20 weeks by

dates. Normal clinical growth until spontaneous labour occurred on 23.4.1971. After 9.25 hours of labour, a vaginal examination was performed as variable deceleration of the fetal heart was occurring with contractions. A thin well effaced cervix which was approximately 8cm dilated was found; a well flexed vertex was judged to be 2+cm below the ischial spines. A diagnosis of cord compression was made and when no change was observed with the patient in the left lateral position, it was decided to expediate delivery. As the cervix was not fully dilated and the head was in the right oblique diameter, it was decided to apply the vacuum extractor.

Delivery of a 3120gm female infant with a five minute Apgar of 8/10 was surprisingly readily achieved. At delivery, the umbilical cord was found to encircle the fetal neck once. The explanation for the cord compression was not obvious to the author. The placenta weighed 566gm and appeared perfectly normal.

Four hours post-delivery the baby was found to have a tachycardia and a rapid respiratory rate. Upon examination faint crepitations were heard at both lung bases and a cardiac flow murmur had developed. A diagnosis of insipient cardiac failure was made and treatment instituted whilst a sample of blood was taken for haematological examination. This revealed a Haemoglobin of 11 grams per cent, a Packed Cell Volume of 39% and a Reticulocyte count of 8%. Because these figures suggested either a chronic blood loss or an acute loss some time prior to delivery, the placenta was re-examined, as were the records of the three amniocenteses. Critical examination

of the placenta revealed no overt evidence of trauma. The liquor at delivery was bloodless as was the liquor on the three occasions of amniocentesis. No fetal cells were ever demonstrated in the maternal circulation.

Following a blood transfusion of packed cells, the baby made a rapid clinical recovery and was discharged fit and well seven days later, having regained its birth weight on the 5th day.

Although no evidence of fetal, umbilical cord or placental trauma was found, and no evidence of a transplacental haemorrhage was found, the last amniocentesis five days prior to delivery must be assumed to be the cause of this neonate's clinical condition. The placenta was found, by means of ultrasound, to be situated in the fundus, so the aetiology becomes even more difficult to explain.

Patient number: 17 L.N.M.P. 1.8.1970

E.D.D. 8.5.1971

This 175cm tall lady weighed 66.9kg at booking at 14 weeks by dates. Her weight gain was 12kg between then and 37 weeks. All clinical parameters were normal until at 34 weeks an External Cephalic Version was attempted for a breech presentation. This failed and was again attempted one week later under sedation (20mg of Diazepam intramuscularly). Consultant opinion sought at this stage was that this patient's obviously powerful abdominal musculature precluded the success of a version and that as a 'primigravid breech' the delivery should be by means of Caesarean section once spontaneous labour had started.

At the time of her last amniocentesis on 21.4.1971, the consultant measuring the biparietal diameter discovered the breech presentation and as the breech did not appear, ultrasonically, to be in the pelvis, attempted an External Cephalic Version. This manouvre was easily accomplished and no pain was experienced by the patient. The fetal heart was clear and regular on auscultation after the version. An uncomplicated amniocentesis was performed some 30 minutes later and as no untoward features had developed an hour later, the patient was allowed home.

The examination of the maternal blood smears showed an absence of fetal cells both before and after the amniocentesis. The placenta had once again been demonstrated, ultrasonically, to have a fundal insertion.

Twelve hours later the patient presented in the labour ward with a history of continuous mild abdominal pain and vaginal bleeding of an hour's duration. On examination the uterus was non-tender and the fetal heart was clear and regular at 136 beats/minute. In view of the history of an External Cephalic Version and an amniocentesis that morning plus the feature of a fundal placenta, a diagnosis of abruptio placentae was made and a lower segment Caesarean section performed. No clotting defect was demonstrated.

At Caesarean section the clinical diagnosis was confirmed as was the site of insertion of the placenta. Upon examining the placenta, approximately 25% of the maternal surface was covered by adherent blood clot.

A 2780gm female infant was delivered with a five minute Apgar score of 9/10.

Post-operatively both baby and mother had uncomplicated courses and were discharged fit and well on the 7th post-operative day.

Obviously an error of judgement was made by the author in performing an amniocentesis within minutes of an External Cephalic Version. Any argument as to which of the two procedures was the cause of the abruption is superfluous. The lesson to be learnt is clear and the author has learnt it. It is further suggested, on the basis of this example, that performances of these two procedures on the same patient should be avoided as far as possible, irrespective of the time interval between them.

4. CLINICAL ASSESSMENT

Clinical assessment of the size of the uterus appears to fall into two categories. Examination of the uterine size early in pregnancy, especially bimanual examination would seem to be the surest means of checking the validity of the patient's dates (Greenhill, 1965; McClure Browne and Dixon, 1970). None of the patients in this study were examined by the author at their initial visit; they were seen in the routine manner at the various booking clinics. It is gratifying to see that there is excellent correlation between uterine size and calculated duration of pregnancy (Table V). This table indicates, however, that our patients in general could conceivably derive even greater benefit if they booked at an earlier stage in their pregnancies, when the chance of error would be reduced even further. This field is obviously that of health education and one which needs attention.

Once the pregnancy gets beyond 20 weeks, the uterine size as an index of the duration of pregnancy becomes unreliable as a result of wide variation in fetal size, indicated by birth weight, brought

about by a multiplicity of factors other than genetic constitution. To expect a reliable answer when measuring one variable such as uterine size against another as variable (the large variation in position of abdominal anatomical landmarks - Beazley and Underhill, 1970), is obviously to expect the impossible. As a generalisation, this method is doubtless applicable, but when an individual patient is considered, extreme caution must be exercised.

When a reliable date of quickening can be obtained from a patient, as was the case in each of the patients in this research group, an index of the duration of pregnancy can be obtained. The duration of pregnancy at which subjective fetal movements were first felt by the patients in this group was 19 (± 2) weeks and compares very favourably with the findings of Kraus and Hendricks (1964) seen in Table VI. The fact that the multigravidae in this latter report had a date of quickening that was two weeks earlier (17 weeks), is probably explained on the basis of previous experience enabling them to interpret what they feel as fetal movements. The impression gained from working with the patients encountered in the Cape Town unit is that the patient who can provide an accurate date of quickening is the exception rather than the rule.

Greenhill (1965) claims the average height of the fundus above the pubis to be 35cm at term, a figure which agrees fairly well with that obtained in this study, although the range in measurement is 33 - 37cm, giving a gestational age of 36+ weeks (Table VII).

McDonald's rule (1908) that "the length in centimetres divided by 3.5 gives the duration of pregnancy in lunar months" is too broad a generalisation to have any application in modern obstetric practice. The statistically significant difference in mean measurements between the normal and 'light-for-dates' pregnancies in this

study may well indicate that this parameter could be used as one of fetal growth in preference to one of gestational age. The value of change in maternal weight gain as an index of fetal growth and, therefore, wellbeing (McClure Browne and Dixon, 1970) is well known. The results of this study suggest that the measurement of the fundal height in centimetres above the pubis might be a practical adjuvant to maternal weight gain patterns in this respect.

The derivation of the uterine volume provides a three dimensional estimation of the size of the uterus. Clinical estimations of the size of the baby (Loeffler, 1967) have been shown to be inaccurate, at the extremes of birth weight, by more than 1lb. (454gm), in over 50% of estimations. As would be expected, measurement of the uterine volume shows a reasonable correlation with birth weight (Figure 20). Better correlation could be expected if the actual uterine volume could be measured and plans are now afoot to attempt such a measurement using ultrasound.

A significant difference is observed between the volumetric measurements of the normal patients and those of the 'light-for-dates' patients. This suggests that this parameter could be used as an index of fetal wellbeing rather than gestational age as placental insufficiency is related to a decreased fetal size and a decrease in the volume of liquor amnii (Beischer et al, 1968).

Hendricks et al (1970) suggest that progressive dilatation of the cervix occurs in an orderly fashion during the last four weeks prior to the onset of labour and the findings in this study corroborate their results. This study was, however, confined to primigravidae and, therefore, no extrapolation to multigravidae can be considered.

5. ULTRASONIC CEPHALOMETRY

This unit is fortunate in that it is one of the two in the country to possess a Diasonograph. Its use in this study has confirmed the value of cephalometry in the assessment of gestational age although, as pointed out by Campbell (1969) the range of values after 30 weeks is large (Figure 23). Segmental analysis of these results indicates that a measurement of more than 92mm is compatible with a gestational age of more than 34 weeks. A measurement of less than 90mm indicates a gestational age of less than 36 weeks in normal pregnancies, but not in the 'light-for-dates' pregnancies, (Figure 26).

The biparietal diameter is unable to predict the birth weight except to say that all fetuses with a measurement of more than 96mm had a birth weight of more than 2,600 grams. A better correlation might have been obtained had the final measurement been obtained nearer the time of delivery. The mean growth rate of the normal infants in this study was just over 1.1mm per week, a figure considerably lower than that obtained by Campbell (1969). The explanation for this is that the means for the first and last measurement were subtracted and the answer divided by the number of weeks. Had the total growth of each infant been meaned and divided by the number of weeks, the answer may well have been higher than 1.1mm/week. Comparing the results in Figure 16 with similar ones (Butler and Alberman, 1969) the neonates in this series do not appear smaller than those of the British Perinatal Mortality Survey and the above is the only explanation for the discrepancy that the author can offer. The accuracy of our Diasonograph was checked before the start of this study so that if the error lies in the actual measurements, it is in subjective undermeasuring by the technician.

The statistically significant difference between the two

groups of patients implies that their serial measurements will indicate those fetuses exhibiting poor intra-uterine growth. Once such a fetus has been identified it is essential to know whether it is in jeopardy or not by assessing its ability to respond to stressful situations. Baillie (1971, personal communication) has recently developed an hypoxic stress test which appears to hold much promise for the future.

With respect to the safety of diagnostic pulsed ultrasound, Donald (1968) states that "hitherto all attempts to measure the physical effects of sonar at the power levels employed in diagnostic work have failed, and so far it has not been found that this technique has caused any harm". This statement is born out by other workers in the fields of both animal experiments and clinical use (Donald and Brown, 1961; Andren, 1964; Willocks et al, 1964; Thompson et al, 1965; Sundén, 1965; Durkan and Russo, 1966; Campbell, 1969). In a survey of 1114 mothers exposed to diagnostic ultrasound, Hellman et al (1970) were unable to demonstrate any increase in the rate of congenital malformations in their offspring.

6. LIQUOR CYTOLOGY

There seems to be considerable agreement amongst authors in this field that false negative fat cell counts do occur after 38 weeks gestation, (Gordon and Brosens, 1967; Sharp, 1968; Chan et al, 1969; Barnett and Nevin, 1970; Lind, 1970; Bishop and Corsen, 1968). The results obtained in this study are in agreement with those of the above workers. False positive fat cell counts were obtained in five of the 47 patients studied i.e. cases numbered 2, 3, 11, 15 and 21 (Appendix E) for which no satisfactory explanation can be forwarded. The 'light-for-dates' fetuses exhibited the same pattern as the normal ones but for the lack of false positive counts. This may, however, be

a reflection of the small number of such cases and not a true difference between the two groups of fetuses. Sharp (1968) and Anderson and Griffiths (1968) found similar results with regard to 'dysmature' fetuses.

Clumping of fat cells occurring near term was first noted by Brosens (1966) and is confirmed in this study. Only two infants exhibited this feature under 36 weeks gestation and then the clumping was of a minor degree. Sixty percent of infants in this study showed this feature after 37 weeks gestation (Figure 27) and there was no difference between the 'light-for-dates' and normal infants in respect of this parameter.

The author suggests that a physiological process of shedding of vernix caseosa, demonstrated to occur after 37 weeks by Brosens et al (1969) is the explanation for this clumping of fat cells as well as for the rise in fat cell count after this time. The fat cell and the polygonal cell would seem to be the same cell (Figures 29 and 30) and to have its origin in the fetal skin. Physiological preparation of the skin for an extra-uterine life necessitates shedding of the protective layer of lipid-laden vernix caseosa, thus exposing the underlying skin and facilitating the exfoliation of cells.

From Figure 7 it can be seen that the polygonal cell is more difficult to count, as individual identification is hampered by the translucent nature of the cell. The general pattern of the rise in polygonal cells with advancing gestation (Figure 29) is similar to that exhibited by fat cells (Figure 28), although comparison of the two cell counts shows a lower polygonal cell count at the same period of gestation. The author suggests that this difference is apparent and not real and is explained by the technical

difficulties of counting the latter as a result of its morphological characteristics. False positive and false negative results similar to those obtained with fat cell counts were obtained, and there was no difference between normal and 'light-for-dates' results. Huisjes and Arendzen (1970) found no false positive results and 14% false negatives. The results in this study do not corroborate their findings, the explanation for which is not apparent.

From Appendix E, it will be seen that in addition to the two samples of liquor amnii not obtained, no results were obtained from a differential cell count on a further four samples. These were from cases numbered 15, 23, 27 and 31. The reason for this was that they were too heavily blood-stained. The blood cells were naturally concentrated with the cells in the liquor amnii as a result of the sample being centrifuged.

The drop in precornified cells and concomitant rise in cornified and anucleate cells found by Lind (1970); Votta et al (1968); Floyd et al (1969); and Bishop and Pollock (1970) are not confirmed in this study. The reason for this was the addition of polygonal cells to the differential cell count in this project. With a rise in polygonal cells occurring after 37 weeks there must of necessity be a fall in the relative numbers of other cells. In view of the agreement between the above authors, a double differential count could have been done in this study; one including and the other excluding polygonal cells. The basal cells recorded in the counts done did not show any significant trend at all and the sighting of amnion cells was a rare occurrence.

The presence or absence of a vagina in the fetus is the basis for the method of sex determination described in Chapter 5, section 6D. The explanation for the cytological pattern would appear

to be an hormonal one. The cytological patterns in smears made from the first specimen of urine voided by the newborn female are virtually identical to the patterns seen in vaginal smears of their mothers taken only hours prior to delivery, (Harrison, V.C., personal communication). The dominant cell in both cases is the intermediate cell.

This finding tends to suggest that a female infant at ± 32 weeks gestation would more often be assigned a gestational age in advance of the known duration of pregnancy than would a male, if a differential cell count which ignored the presence of polygonal cells were used. The accuracy of this method of pre-natal sex determination in the early stages of pregnancy was not studied in this research project, but in a publication after the completion of this work, Arendzen and Huisjes (1971) suggest that early differentiation can be made.

7. LIQUOR BIOCHEMISTRY

The source of urea in the liquor amnii is thought to be the fetal urine (Lind, 1970). He suggests that increased renal function leads to a progressively more hypotonic urine, explaining the fall in osmolality and sodium and the rise in urea and creatinine concentrations.

On the basis of this theory, the rise in urea, creatinine and true creatinine concentrations in the liquor amnii with increasing gestation (Figures 37, 40 and 42) is explained. The wide range in concentrations for specific weeks of pregnancy is not as easily explained. There would appear, from the results obtained in this study, to be very little correlation between the concentrations of these three chemicals and the ultimate birth weight. As only one infant was found to be depressed, as evidenced by an Apgar score of 6/10, no correlation can be made with fetal wellbeing or state at

delivery.

It must remain conjectural whether the answers lie in the total amount of these substances rather than their concentrations.

Of the biochemical parameters, the best for estimating gestational age would appear to be the true creatinine and the true creatinine difference between maternal serum and liquor amnii (Figures 42 and 44). As judged by the correlation coefficients, the order in which the other biochemical parameters best relate to gestational age is total creatinine, liquor-maternal urea difference and liquor urea (Figures 40, 39 and 37).

That the concentration of bilirubin in the liquor appears unable to act as an index of gestational age might be attributable to one or all of the following reasons:

i. it is in fact not an index of gestational age

ii. the method of estimation, with an experimental error of ± 6% was the wrong one. It might have been measuring any one or combination of several pigments known to occur in normal amniotic fluid, including bilirubin, biliverdin, coproporphyrin III and urobilinogen (Brown, 1970).

iii. the variable exposure to light that each sample invariably had may have resulted in differing degrees of photo-oxidation.

The author suggests that further studies be done, where possible, upon the analysis of liquor amnii for the presence of bilirubin by means of spectrophotometry. It is further suggested that this be done as soon after amniocentesis as possible and that the liquor be transported in a light-proof container.

8. RADIOLOGY

The subject of the hazards of exposure to ionizing radiation during the pre-natal period have already been discussed (Chapter 3, section 10), and will not be covered here. Suffice to say that the radiation dose to the fetus must be kept as low as possible, and that the later in pregnancy that an X-ray be taken, the better. An X-ray prior to 36 weeks is less helpful than one thereafter, accepting that often the duration of pregnancy is unknown and is the indication for the investigation.

Of the various methods of radiological assessment of fetal maturity (Hartley, 1957; Stockland and Marks, 1961; Brandfass and Howland, 1967) it was decided to use only the presence or absence of the ossification centres around the knee joint. Dee et al (1966) examined twins soon after delivery for the presence and size of these centres. Radiological conditions were ideal and all sets of twins were presumed to be the same gestational age. Little correlation between the ossification centres of each pair was found, leading them to conclude that these centres were a poor index of gestational age. A similar conclusion was reached by Cope and Murdoch (1958). Add to these findings the demonstration by Scott and Usher (1964) that fetuses subjected to 'intra-uterine malnutrition' show retarded ossification of the epiphyses, and only one conclusion can be reached. Except when the pregnant patient has diabetes mellitus, an X-ray of the fetus which demonstrates the presence of one or both of the epiphyseal centres at the knee can be interpreted as one of a fetus of 36 or more weeks gestational age. The converse is not only wrong but may well be dangerous to the fetus with regard to its future survival.

The results of X-ray examination of the fetuses in this project show that those fetuses X-rayed after 35 completed weeks, but

before the completion of the 36th week, tended to be over-assessed by the radiologist, the mean radiological age being 36.8 weeks. Those X-rayed between the 36th and 37th completed week were assigned a mean age of 36.3 weeks but the range is from 34 to 38.5 weeks (Figure 46). Those X-rayed after the 37th completed week were given a mean radiological age of 36.4 weeks and here the range was 3 weeks. Giving the radiologist no latitude at all, from Figure 46 it can be seen that 51% of fetuses were over-assessed and 36.2% were under-assessed. It is interesting to note that all but one 'light-for-dates' fetus were under-assessed.

There was no statistically significant difference between the radiological assessments of the two race groups i.e. the two radiographic techniques were equally efficacious.

9. DISCRIMINANT ANALYSIS

The results of the first analysis done revealed that the best parameter of gestational age was the clinical assessment of the height of the fundus. At this stage the author requested that the entire analysis be repeated with this parameter excluded. The result of this second analysis are given in Chapter 5, section 10.

As has been explained (Chapter 5, section 10) the discriminant functions derived from the results of this study, should be regarded as a method of classification of a pregnancy into one of three time groups, rather than as a list of variables which are related to the age of a fetus.

Comparing tables XVI and XVII, it is obvious that the discriminant functions given in Table XIV are superior to those given in Table XV.

After the completion of this work, two separate scoring systems were published (Lind and Billewicz, 1971; O'Leary and Bezjian,

1971). From the results of this investigation it has not been possible to derive a similar scoring system. With the aid of a simple calculator it is, however, relatively simple to use the discriminant functions given in Table XIV. The choice of these discriminant functions was governed by the fact that they were derived after elimination of the factor of dependence, and that they were the better of the two sets of functions so derived.

To use them, figures must be obtained for all five variables. These must be substituted; ultrasonic biparietal diameter measurement for y_1, clumping of fat cells for y_2, etc., and the calculation completed. This must be done for all three functions. That function which results in the highest figure signifies into which time group the patient belongs.

A composite picture can thus be built by consideration of all available data concerning a patient whose duration of pregnancy must be known i.e. clinical evaluation of the height of the fundus and/or the estimated size of the fetus, radiological examination and special investigations including use of the discriminant functions.

From a combination of these parameters, the author believes, on the basis of this study, that an accurate assessment of the gestational age can be made in normal pregnancies. The next step is obviously to test these discriminant functions in pregnancies other than normal but this is not within the scope of this study.

Following the biochemical and cytological estimations performed in this study, all remaining liquor was returned to the deep freeze to be stored at $-20^{o}C$. The work of Gluck et al (1971) has prompted further analysis of this liquor amnii. Work is now proceeding on the assessment of pulmonary maturity from examination of the liquor amnii. A sudden rise in the concentration of the

phospholipid Lecithin is claimed by Gluck et al, to occur at 35 weeks gestation, and to herald the development of pulmonary surfactant. This substance is believed to be absent from the lungs of neonates with severe respiratory distress syndrome. At the time of writing, the author has insufficient results to draw any conclusions, but if Gluck et al, are correct, this estimation will not only be an additional parameter of gestational age, but a valuable adjunct in the assessment of functional maturity of a fetus.

CHAPTER 7

CONCLUSIONS

1. The date of the last normal menstrual period provided by any patient in our unit will be incorrect by more than two weeks in over 33% of cases. A further 5.1% of our European patients and 17.1% of our non-European patients are unable to provide any such date at all. Elective termination of pregnancy accounts for one in every seven deliveries in our unit and it is, therefore, necessary to balance the advantage of early termination (either by induction of labour or elective Caesarean section) against the risk of delivering an infant of shorter gestation than anticipated. Reliable methods of assessing gestational age are, therefore, of paramount importance to the fetus.

2. Using a strict criterion of one standard deviation below the mean weight for age, 'light-for-dates' infants were born of 17% of clinically normal pregnancies in this study. From the results obtained from these pregnancies, it is obvious that the slow growth rate exhibited by these fetuses could have been identified during the antenatal period, had the results of the investigations been interpreted as they were obtained.

3. The criteria by which the patients in this study were selected, are believed to have reduced the influences of socio-economic factors in the two ethnic groups studied, to the same level.

4. Early clinical evaluation of uterine volume by bimanual examination is an important parameter of the duration of pregnancy. The patients in this study booked at a mean gestation of 16 weeks. School and community education is needed to emphasise the benefits of

early booking. The keeping of a menstrual calander by women exposed to the possibility of pregnancy also needs emphasis.

5. The date of quickening provides an approximate index of the duration of pregnancy. In this study, quickening occurred at 19 (± 2) weeks.

6. Clinical assessment of the height of the uterine fundus relative to abdominal anatomical landmarks, although accurate as a parameter of gestational age in this study, was probably influenced by observer bias. No conclusions can be drawn from this parameter.

7. Measurement in centimetres of the height of the uterine fundus above the symphysis pubis shows a wide range, even in the small number of patients in this study. It would appear, however, to provide a clinical measurement of retarded fetal growth, although no more than an index of suspicion can be inferred from this parameter.

8. The uterine volume, by the nature of its derivation, is only a very approximate measurement. It does appear to provide not only a rough guide to fetal weight, but a more reliable index of fetal growth than the measurement of the fundal height alone. This parameter is now incorporated into the routine examination of all antenatal patients in our unit, not as a parameter of gestational ageing, but as a serial monitor of fetal growth.

9. 'Taking up' of the primiparous cervix appears to occur from 36 weeks onwards, and as such provides a rough guide that delivery will probably take place within four weeks of this occurring. The finding of a long, thick and undilated cervix in a primiparous patient does not mean, however, that the spontaneous onset of labour is more than four weeks away.

10. Wide biological variation was found in each of the parameters measured by means of non-clinical methods.

11. Ultrasonic biparietal diameter measurements enable an approximate gestational age to be derived but the range in this study for any measurement over 90mm is just over five weeks. Segmental analysis would seem to be of value at the 34 - 36 week period of gestation. At present, any patient who is found at her first visit, to have a discrepancy between the expected and actual size of her uterus, is referred for an ultrasonic biparietal diameter measurement. It is believed that this measurement, as a parameter of gestational age, is more accurate prior to 30 weeks gestation, than afterwards.

12. Serial ultrasonic biparietal diameter measurements provide a precise index of fetal growth.

13. Birth weight could not be predicted from knowledge of the biparietal diameter measurements obtained in this study.

14. Amniocentesis is a procedure not devoid of risk to both fetus and mother. It is recommended that amniocentesis not be performed for research purposes only, and that a definite indication be present before it is contemplated.

15. Placentography should precede every amniocentesis. In this unit, routine ultrasonic placentography is performed prior to all amniocenteses.

16. Amniocentesis should not be performed except under full sterile precautions. In this manner the danger of intra-uterine infection is minimized.

17. Experience in the technique of amniocentesis would appear to be important for the reduction of complications. It is, therefore, recommended that the inexperienced should not undertake this procedure without the guidance of someone with experience.

18. Fat cell counts appear to correlate well with gestational age when there are more than 10% fat cells present. The number of false negative counts in this study is much larger than the number of false positives. From this study it would appear that a fat cell count in excess of 10% is suggestive of a gestational age of 35 weeks or more. A fat cell count of less than 10% is unhelpful as an index of gestational age.

19. The method of staining liquor amnii with Nile Blue sulphate evolved for this study would appear to be a highly satisfactory one.

20. Fat cells appear to be keratinized squamous cells coated with a layer of lipid material, rather than cells containing intracellular lipid as was suggested by earlier workers.

21. 'Light-for-dates' fetuses do not differ significantly from their normal counterparts in respect of fat cell counts in this study.

22. Clumping of fat cells is a feature suggestive of a gestational age of 37 weeks or more. The absence of this feature is not necessarily an index of a gestational age of less than 37 weeks. This characteristic needs to be quantitated in some manner to make it more useful. Because counting of the cells in a clump is very often impossible, the author knows of no other way than the use of a micrometer. This was not done in this study. The aggregations of cells are likely to be disrupted with the passage of liquor amnii through the needle at amniocentesis, as well as during the staining process. It is, therefore, suggested that the aggregations which survive these

two hazards, are a true reflection of the intra-uterine occurrence thereof.

23. The author suggests that the clumping of cells is a result of physiological shedding of the vernix caseosa in preparation for the extra-uterine environment.

24. Polygonal cells are believed to be fat cells denuded of their extra-cellular lipid by the staining process. That fewer polygonal cells than fat cells are found in the same specimen of liquor, is considered to be apparent and not real. Centrifuging the liquor amnii results in the production of large aggregations of cells and the translucent nature of the polygonal cells makes individual counting difficult under these circumstances.

25. A differential cell count which includes the counting of polygonal cells, as was used in this study, provides very little index of gestational age. A rise in polygonal cells must be balanced by a proportionate drop in the other cell types. The addition of polygonal cells to the differential count does not, however, allow for quantitative evaluation of changes occurring in the other cell types with advancing gestation, if changes in fact occur.

26. Antenatal sex determination after 30 weeks of pregnancy can be achieved simply and with a high degree of accuracy using the cyanophilic cell count on liquor amnii. Passage of urine over the vestibule of the vagina, from which the majority of cyanophilic staining intermediate cells originate, is thought to be the mechanism whereby these cells get into the liquor amnii. The high cyanophilic cell counts, therefore, indicate the presence of a vagina.

This method of sex determination has not been tested in

early pregnancy, but if fetal urine starts to contribute to the volume of liquor amnii from 14 - 16 weeks onwards, it is expected that a similar situation as is found after 30 weeks, would be found from ± 16 weeks onwards. This may prove a useful adjuvant to the method already in widespread use i.e. the identification of the sex chromatin or Barr body in the nuclei of cells. A disadvantage of this latter method, is that a sufficient number of unimpaired nuclei are not always found in the smear.

27. Knowledge of the liquor volumes would have allowed total amounts of the biochemical constituents to be calculated. These values may have provided better indices of gestational age than the concentrations measured in this study.

28. Assessed individually, the biochemical parameters measured in this study afford little more than an approximation of gestational age to be derived. The true creatinine would seem to be a better parameter than the others, both in respect of its actual concentration in the liquor and the difference between it and the maternal serum concentration. True creatinine is now routinely estimated for the Gestational Ageing Service in preference to the total creatinine.

29. None of the biochemical parameters measured in this study appear to be influenced by the factors which influenced fetal growth and led to the 'light-for-dates' infants.

30. Summation of parameters is able to provide a far more accurate index of gestational age than any single parameter. Although the actual week of gestation, if known, is a more specific figure with which to deal, it was felt that three-week time groups were clinically as applicable. These three time groups were selected at the start of

this project, when it was believed that the biological range for any single parameter at any single week in pregnancy, would preclude the assessment of gestational age in specific weeks.

31. The discriminant functions derived from the discriminant analyses, allowed correct classification into the three time periods selected, of 89% of the pregnancies studied.

32. Radiological assessment of gestational age would appear from this study to be of limited value only. It can be inferred that the radiological presence of a distal femoral epiphysis is indicative of a pregnancy that has reached 36 weeks duration. The absence of this ossification centre does not necessarily indicate a gestational age of less than 36 weeks.

33. The under-assessment of all but one of the 'light-for-dates' fetuses, indicates delayed ossification of the epiphyseal centres round the knee joint in these fetuses, even when mild degrees of growth retardation are present.

34. By the physical and neurological examination of a neonate, the paediatrician is able to derive a gestational age that is within two weeks of the calculated age of that neonate at delivery.

35. Now that the physiological changes occurring with advancing gestation in normal pregnancies, with respect to the parameters assessed in this study, are known, pathological states of pregnancy need similar investigation. Some of the effects of conditions such as diabetes mellitus, essential hypertension, chronic renal disease etc., are well established. What is not known, however, is what affects these conditions have on the majority of parameters measured in this study.

The author plans a similar study on groups of patients with such conditions, in an attempt to define any significant deviations from the normal. It is conceivable that to assess the gestational age, different parameters need to be measured in different pathological states of pregnancy.

36. The author considers that apart from the knowledge of the duration of any pregnancy, indices of fetal wellbeing and fetal growth are essential for the management of complicated pregnancies. It is also essential that some method of prediction of the condition at birth be evolved. Hypoxic stress tests have so far provided an accurate index of fetal wellbeing but the correlation with condition at birth could be improved upon.

37. Hyaline membrane disease is the most lethal hazard of prematurity, and as such, an index of pulmonary maturity prior to induction or Caesarean section is required. The author is at present involved in the quantitative estimation of phospholipid fractions in the liquor amnii of the patients in this study, in the hope that such an index can be found.

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APPENDIX A

The consent form used to obtain consent from both husband and wife for the incorporation of the latter into the study group.

We, the undersigned Husband and Wife, hereby state that the nature, scope and consequences of Amniocentesis and Ultrasonic Examination have been explained to us by Dr. Bennett and we appreciate and accept any risks associated with, and the consequences arising out of such procedures.

SIGNED: .....(Husband)

.....(Wife)

WITNESSES:

1. .....

2. .....

DATE: .....

Ons die ondergetekendes, man en vrou, verklaar hierby dat die aard, omvang en gevolge van Amniosentese en Ultrasonies ondersoek deur Dr. Bennett aan ons verduidelik is. Ons besef en aanvaar enige risiko's verbonde aan en gevolge wat mag voortspruit uit sodanige ingrepe.

GETEKEN: .....(Man)

.....(Vrou)

GETUIES:

1. .....

2. .....

DATUM: .....

The above is a copy of the consent form signed by both husband and wife prior to the wife's inclusion in the research programme.

APPENDIX B

The forms in this appendix are facsimiles of those used to record the data obtained for each patient in the study group.

PROGRAMME NO: NAME: FOLDER NO:

AGE: HUSBAND'S OCCUPATION: BLOOD GROUP:

BOOKING: HEIGHT: cms. WEIGHT: kg.

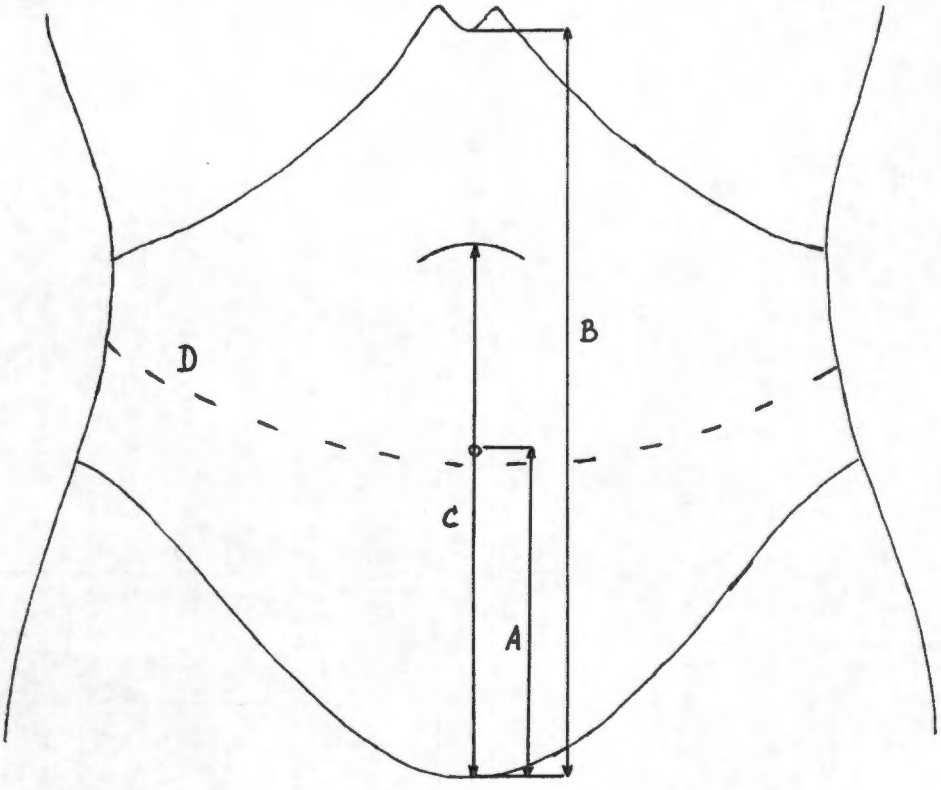
L.N.M.P.: CYCLE / "PILL" YES/NO

F.M.F.: SURE | UNSURE

INITIAL P.V. DATES: /52 UTERINE SIZE: /52

DATE	Duration of pregnancy in weeks	Weight kg	B.P.	Urinalysis	H.O.F. clinically	H.O.F. cms	STATE OF CERVIX

PROGRAMME NO: NAME: FOLDER NO:



A. Symphysis to Umbilicus

B. Symphysis to Xiphisternum

DATE	C. (H.O.F. in cms)	D. (Max. Girth in cms)

Comprehensive Recording Form

for all Special Investigations.

PROGRAMME NO:

Completed weeks	30	31	32	33	34	35	36	37	38	39	40
Maternal urea											
Maternal true creatinine											
Maternal bilirubin											
Liquor urea											
Liquor creatinine											
Liquor true creatinine											
Liquor bilirubin											
Ultrasonic B.P.D.											
X-ray maturity											
Complications of amniocentesis											
Uterine volume											
Pre. Post Maternal Kleihauer											
Weight in kg.											

											H.O.F. clinically
											State of cervix
											Macroscopic appearance
											Microscopic blood
											Fat globules
											Blue squames
											Yellow anucleates
											Fat cells
											Clumping of fat cells
											Polygonal cells
											Anucleate cells
											Superficial cells
											Intermediate cells
											Basal cells
											Amnion cells

PROGRAMME NO: NAME: FOLDER NO:

L.N.M.P.:

DATE OF DELIVERY: .˙. ACTUAL GESTATIONAL AGE:

DELIVERY:				
	S.V.D.	YES	NO	INDICATION
	INSTRUMENTAL	YES	NO	
	L.U.S.C.S.	YES	NO	

INFANT: BIRTH WEIGHT: Grams

SEX: F M

HEAD CIRCUMFERENCE: Centimetres

LENGTH: Centimetres

APGAR: 5 min.: /10

PLACENTAL WEIGHT: (Minus membranes and cord) = Grams

PAEDIATRIC GESTATIONAL AGE = Weeks

COMMENT:

EXAMINATION OF INFANT AT 6 WEEKS:

APPENDIX C

The following tables list the results of repeated estimations upon known concentrations of the various substances. The standard solutions were made up to arbitrary concentrations within the expected range so as to be able to check on the accuracy and reproducability of the methods being used.

TABLE I

UREA CONTROLS

Standard Known Value:	20mg%	30mg%	40mg%
	19.5	30.6	39.7
	18.9	30.3	40.5
	19.4	30.8	40.5
	19.5	30.3	39.7
	19.5	30.3	39.7
	19.4	30.3	39.7
	19.5	29.9	39.7
	19.5	30.6	39.7
	19.6	30.2	39.7
	19.3	30.4	40.4
	19.5	30.3	40.3
	19.8	30.6	40.5
	19.4	30.2	39.7
	19.5	30.3	39.9
	19.6	30.4	39.7

TABLE II

CREATININE CONTROLS

Standard Known Value:	0.8mg%	1.6mg%	2.8mg%
	0.81	1.44	2.61
	0.84	1.43	2.62
	0.76	1.44	2.51
	0.76	1.44	2.51
	0.81	1.43	2.51
	0.79	1.45	2.51
	0.84	1.44	2.51
	0.81	1.43	2.53
	0.79	1.45	2.52
	0.79	1.48	2.51
	0.81	1.44	2.51
	0.84	1.43	2.52
	0.79	1.44	2.51
	0.79	1.45	2.53
	0.81	1.43	2.51

TABLE III

TRUE CREATININE CONTROLS

Standard Known Value:	0.8mg%	1.6mg%	2.0mg%
	0.79	1.57	1.98
	0.80	1.58	2.02
	0.81	1.60	1.94
	0.81	1.57	1.96
	0.80	1.60	1.96
	0.81	1.61	2.02
	0.79	1.60	1.94
	0.79	1.60	2.02
	0.81	1.58	2.03
	0.80	1.60	2.04
	0.81	1.59	2.02
	0.79	1.61	2.03
	0.81	1.60	1.98
	0.79	1.61	1.98
	0.79	1.61	1.96

TABLE IV

BILIRUBIN CONTROLS

Standard Known Value:	0.50mg%	0.25mg%
	0.46	0.24
	0.48	0.25
	0.52	0.24
	0.56	0.26
	0.48	0.25
	0.47	0.24
	0.51	0.24
	0.57	0.26
	0.53	0.24
	0.54	0.25
	0.48	0.25
	0.49	0.26
	0.51	0.24
	0.43	0.24
	0.44	0.26

TABLE V

THE EFFECT OF FREEZING UPON THE UREA, CREATININE AND TRUE CREATININE CONCENTRATIONS IN LIQUOR AMNII

Constituent Estimated	Sample Number	Results	
		Fresh mg%	Frozen mg%
Urea	1	32.1	33.0
	2	34.4	33.9
	3	36.2	35.8
	4	41.2	41.0
	5	37.7	37.8
Creatinine	1	1.98	2.11
	2	1.89	1.94
	3	2.56	2.39
	4	2.32	2.36
	5	2.91	2.87
True Creatinine	1	1.77	1.71
	2	1.74	1.76
	3	2.21	2.28
	4	2.16	2.22
	5	2.53	2.45

APPENDIX D

Photostat copies of the scoring systems for both neurological and external criteria described by Dubowitz et al (1970), for the paediatric assessment of gestational age. These scoring systems were used on the infants born to the patients in this study.

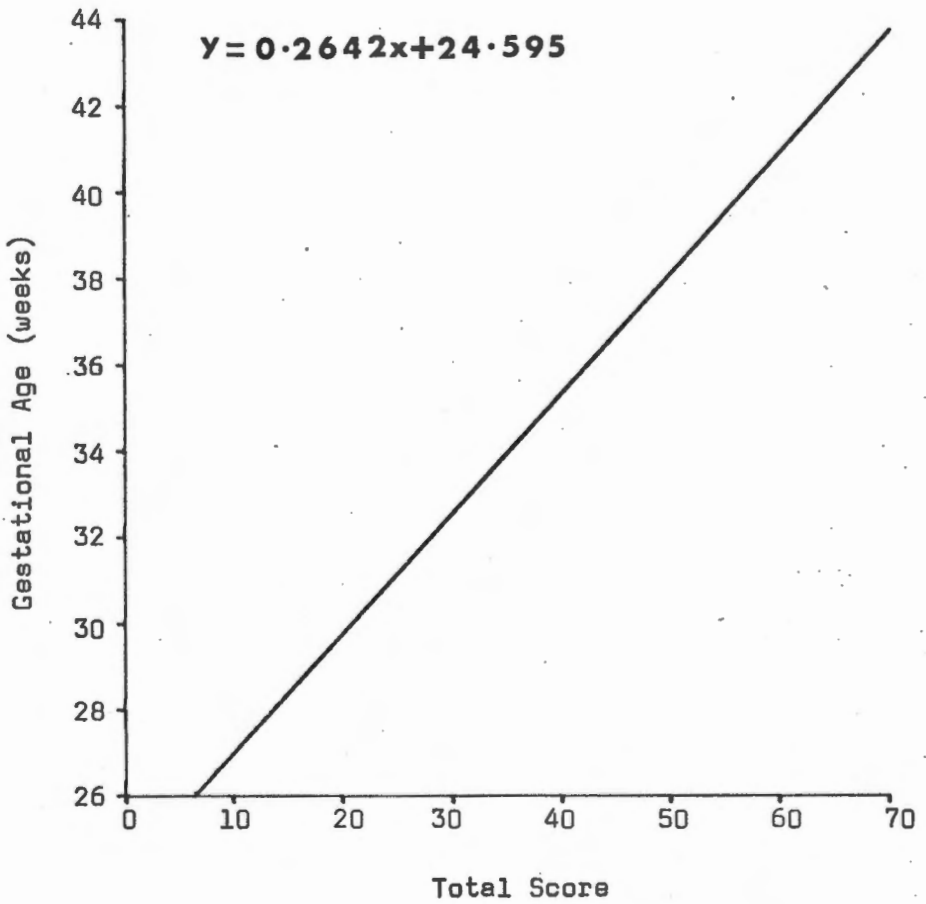
NEUROLOGICAL SIGN	SCORE					
	0	1	2	3	4	5
POSTURE						
SQUARE WINDOW	 90°	60°	45°	30°	0°	
ANKLE DORSIFLEXION	 90°	75°	45°	20°	0°	
ARM RECOIL	 180°	90-180°	<90°			
LEG RECOIL	 180°	90-180°	<90°			
POPLITEAL ANGLE	 180	160°	130°	110°	90°	<90°
HEEL TO EAR						
SCARF SIGN						
HEAD LAG						
VENTRAL SUSPENSION						

Scoring System for Neurological Criteria

External sign	*Score**				
	0	*1*	*2*	*3*	*4*
Edema	Obvious edema of hands and feet; pitting over tibia	No obvious edema of hands and feet; pitting over tibia	No edema		
Skin texture	Very thin, gelatinous	Thin and smooth	Smooth; medium thickness. Rash or superficial peeling	Slight thickening. Superficial cracking and peeling especially of hands and feet	Thick and parchment-like; superficial or deep cracking
Skin color	Dark red	Uniformly pink	Pale pink; variable over body	Pale; only pink over ears, lips, palms, or soles	
Skin opacity (trunk)	Numerous veins and venules clearly seen, especially over abdomen	Veins and tributaries seen	A few large vessels clearly seen over abdomen	A few large vessels seen indistinctly over abdomen	No blood vessels seen
Lanugo (over back)	No lanugo	Abundant; long and thick over whole back	Hair thinning especially over lower back	Small amount of lanugo and bald areas	At least ½ of back devoid of lanugo
Plantar creases	No skin creases	Faint red marks over anterior half of sole	Definite red marks over > anterior ½; indentations over < anterior ⅓	Indentations over > anterior ⅓	Definite deep indentations over > anterior ⅓
Nipple formation	Nipple barely visible; no areola	Nipple well defined; areola smooth and flat, diameter < 0.75 cm.	Areola stippled, edge not raised, diameter < 0.75 cm.	Areola stippled, edge raised, diameter > 0.75 cm.	
Breast size	No breast tissue palpable	Breast tissue on one or both sides, < 0.5 cm. diameter	Breast tissue both sides; one or both 0.5 - 1.0 cm.	Breast tissue both sides; one or both > 1 cm.	
Ear form	Pinna flat and shapeless, little or no incurving of edge	Incurving of part of edge of pinna	Partial incurving whole of upper pinna	Well-defined incurving whole of upper pinna	
Ear firmness	Pinna soft, easily folded, no recoil	Pinna soft, easily folded, slow recoil	Cartilage to edge of pinna, but soft in places, ready recoil	Pinna firm, cartilage to edge; instant recoil	
Genitals Male	Neither testis in scrotum	At least one testis high in scrotum	At least one testis right down		
Female (with hips ½ abducted)	Labia majora widely separated, labia minora protruding	Labia majora almost cover labia minora	Labia majora completely cover labia minora		

Scoring System for External Criteria

* If score differs on two sides, take the mean.



Graph for reading gestational age from total score

APPENDIX E

In this appendix appear all the results from this investigation. The letters D.O.P. are an abbreviation of Duration of Pregnancy, and signify the calculated gestational age at the time when the figure was obtained.

INFANT PARAMETERS

Case No.	D.O.P. at Delivery (weeks)	Birth Weight (g)	Length (cm)	Paediatric ageing (weeks)
1	40.43	2893	48	40.5
2	38.71	3250	55	38.8
3	40.00	3430	53	40.8
4	40.00	2438	48	39.8
5	40.14	4139	50	40.3
6	38.00	2833	54	37.6
7	38.71	3120	52	39.2
8	41.43	3402	53	40.6
9	37.86	3458	53	40.0
10	39.57	2500	50	40.2
11	38.14	2950	50	38.8
12	41.57	2807	50	41.6
13	37.71	2069	51	38.1
14	38.57	2693	48	39.0
15	40.57	3063	50	40.5
16	38.57	3000	52	38.2
17	37.57	2780	48	37.8
18	39.57	2550	47	40.0
19	39.43	3515	53	40.0
20	40.57	3750	53	41.0
21	40.86	3771	56	40.3
22	40.29	2930	54	41.0
23	40.00	3969	53	40.4
24	37.14	3350	54	37.8
25	37.57	3092	54	38.6
26	39.57	3050	56	39.0
27	39.43	3771	50	40.0
28	39.57	3350	52	39.9
29	40.57	2486	45	39.5
30	38.71	3175	55	39.4
31	40.86	3969	57	40.3
32	38.29	2608	52	38.2
33	37.29	2865	54	37.4
34	38.71	2950	52	40.0
35	38.71	3006	50	39.4
36	39.00	3289	57	38.8
37	41.29	2950	48	40.6
38	36.86	3050	55	37.6
39	41.00	3402	50	40.4
40	40.57	3005	51	40.2
41	41.43	3288	50	40.8
42	38.71	2750	49	39.2
43	39.00	3120	51	39.7
44	40.43	3450	58	40.1
45	39.29	2463	49	39.2
46	37.71	2410	49	37.6
47	38.43	2180	46	38.6

STATE OF CERVIX

Case No.	D.O.P.	Grade	D.O.P.	Grade	D.O.P.	Grade
1	31	1	34	1	38	2
2	32	2	35	2	37	3
3	32	1	35	1	38	2
5	32	1	35	1	37	1
6	31	1	35	2	37	4
7	33	1	36	1	38	2
8	31	1	34	1	37	1
9	32	1	35	2	37	3
11	31	1	34	1	37	1
14	32	1	34	1	37	2
15	32	1	35	1	38	2
16	32	1	34	1	37	1
17	33	1	35	1	37	1
19	31	1	35	1	36	2
20	32	1	35	1	37	1
21	32	1	35	1	37	1
22	32	1	35	1	37	1
23	32	1	35	1	37	3
24	33	1	35	1	37	3
25	31	1	34	1	37	3
26	32	1	35	1	37	1
27	32	1	34	1	36	2
28	32	1	36	1	37	1
30	32	1	35	1	37	2
31	31	1	34	1	37	1
32	32	1	35	1	37	3
33	31	1	34	2	-	-
34	31	1	35	1	37	2
35	32	1	35	1	37	1
36	32	1	35	1	37	2
37	31	1	34	1	37	1
38	32	1	35	1	-	-
39	31	1	35	1	37	1
40	32	1	35	1	37	2
41	32	1	35	1	37	1
42	32	1	35	1	38	2
43	31	1	36	1	39	1
44	32	1	36	1	37	1
46	32	1	35	2	37	3
4	31	1	35	1	37	2
10	32	1	35	1	38	3
12	31	1	34	1	37	1
13	33	1	35	1	37	1
18	32	1	35	1	37	3
29	32	1	35	1	37	2
45	32	1	35	1	38	2
47	32	1	35	1	38	2

Uterine Volume Measurements for each Patient

CALCULATED UTERINE VOLUME (cm^3)

Case No.	DURATION OF PREGNANCY IN WEEKS										
	30	31	32	33	34	35	36	37	38	39	40
1	3845		4182		4908		5429		6087	5814	
2		4405		5410		5797		6232	6232		
3		4022		5196	6356	6280	6532	6600	6779	7033	
4	2547	2666		2989		3080	3511	3698	3370	3720	
5		4829		5958	6124		6327	7181	7594	7634	
6	3545	3846		4230	4512		5033	5007			
7			3895		4054			4752	5745		
8		3645		4339		4704	4882	5191	5860		6570
9	3552	3871	4048		4898		4925				
10	3950			4227		4680	5659	5354			
11		4733	5142		5905	6044	5845				
12		4131		4203	3936		4843		5590	5950	5950
13		3568	3655		4763	4817	4672				
14			4844		5047		5861	5693	6158		
15	2788		3138			4179	4314	5357	5357	5765	
16		3615		4478		4529	5637	5700			
17				4776	4562		5629				
18		2677		3225		3916		4383		5412	
19	4428		5358			5439	6088	7248	7403	7164	
20		3297		4612	5114		6044	6422	6191	7140	
21		3714		4961			5354	6599	6389	6324	6620
22		2671		3232		3636	4155	4161	4389	4207	4631
23				5314		5958	6119	6569	6587	6429	
24			3261		4733		5429				
25	4680		5745	6088		6149	6179				

26	2788		3391		3847		5205	5574		5590	
27		5060		5487		6013	5982	6565	7219		
28	2990	3545				5033		5590	5458	5635	
29	2076	2461				3157	3633	3512	4115		
30		3590		4685	5028		5297	5480	5297		
31	3892		4829		5243	5429	5487	6119	6323	6884	6027
32	2858	3263		4442	4537		5354		5382		
33	3106		3893			4682	5033				
34	3007		3722		3964		4826	4741	4682		
35	2941		2941		3546		4466	4466	4611		
36		3490		4060	4107		4672	5171	5699		
37		2548		3741	3420	4026		4442		4611	4513
38	3633	3754		4784	4636		4860				
39	3568		3712		4326		5202		6080	5987	6997
40		3060		4117	4981	5030	5917	5629	6032	6032	
41	2971		3273		3812		4275	4713	4908	5084	5327
42	2722		3489		3489		4278	4639			
43	3682		4316		4936		6027	6804	6804		
44	3782		4758		4369	5169	5458	5738	5354	5966	6599
45		2113		2671		3532		3802	3891	4428	
46		2889		3193		4160	4466		4481		
47	2858	3694		4204		4835	4109	4339	4339		

ULTRASONIC BIPARIETAL DIAMETER

Case No.	D.O.P.	mm.	D.O.P.	mm.	D.O.P.	mm.
1	31	82.5	34	85.7	38	93.0
2	32	-	35	94.0	37	96.1
3	32	82.4	35	93.7	38	100.0
5	32	88.3	35	93.1	37	97.7
6	31	85.8	35	89.6	37	93.7
7	33	88.7	36	93.3	38	95.1
8	31	83.9	34	86.7	37	92.0
9	32	91.0	35	94.3	37	95.6
11	31	88.0	34	91.1	37	93.2
14	32	86.5	34	92.3	37	94.3
15	32	83.4	35	91.1	38	94.1
16	32	87.5	34	91.3	37	96.8
17	33	82.0	35	86.0	37	91.7
19	31	86.4	35	97.5	36	100.0
20	32	90.1	35	97.6	37	99.6
21	32	91.6	35	95.6	37	100.0
22	32	81.6	35	88.5	37	95.7
23	32	88.3	35	94.4	37	97.5
24	33	86.9	35	93.1	37	97.6
25	31	83.9	34	90.6	37	96.4
26	32	87.8	35	92.1	37	96.9
27	32	91.5	34	96.4	36	101.0
28	32	88.2	36	96.6	37	98.0
30	32	91.4	35	96.7	37	100.0
31	31	91.7	34	95.8	37	98.7
32	32	88.9	35	94.7	37	97.0
33	31	86.1	34	94.9	-	-
34	31	90.0	35	91.7	37	96.1
35	32	85.0	35	97.2	37	102.0
36	32	89.6	35	93.9	37	98.3
37	31	86.6	34	95.4	37	101.0
38	32	89.1	35	95.6	-	-
39	31	83.2	35	92.1	37	96.1
40	32	86.4	35	91.0	37	92.6
41	32	89.8	35	96.4	37	98.8
42	32	87.3	35	91.5	38	97.6
43	31	89.0	36	93.9	39	97.6
44	32	89.0	36	94.7	37	99.1
46	32	89.0	35	94.5	37	95.2
4	31	80.0	35	87.2	37	91.0
10	32	87.3	35	91.0	38	94.7
12	31	76.0	34	82.7	37	88.7
13	33	85.9	35	87.2	37	91.0
18	32	77.5	35	82.7	37	88.7
29	32	78.5	35	84.7	37	92.8
45	32	88.1	35	93.2	38	95.1
47	32	83.7	35	87.3	38	92.1

FAT CELLS

Case No.	D.O.P.	%	D.O.P.	%	D.O.P.	%
1	31	0	34	0	38	55
2	32	28	35	28	37	29
3	32	0	35	12	38	11
5	32	0	35	2	37	49
6	31	1	35	1	37	0
7	33	0	36	2	38	22
8	31	0	34	3	37	3
9	32	1	35	2	37	27
11	31	1	34	18	37	32
14	32	1	34	1	37	3
15	32	1	35	-	38	15
16	32	1	34	1	37	31
17	33	1	35	1	37	62
19	31	1	35	8	36	6
20	32	1	35	4	37	28
21	32	1	35	2	37	5
22	32	2	35	5	37	6
23	32	1	35	8	37	8
24	33	1	35	3	37	13
25	31	1	34	3	37	34
26	32	1	35	2	37	2
27	32	1	34	1	36	1
28	32	1	36	3	37	12
30	32	4	35	9	37	25
31	31	1	34	3	37	7
32	32	1	35	5	37	7
33	31	1	34	6	-	-
34	31	4	35	4	37	6
35	32	1	35	3	37	10
36	32	8	35	10	37	25
37	31	1	34	1	37	7
38	32	1	35	15	-	-
39	31	1	35	5	37	7
40	32	5	35	3	37	35
41	32	7	35	21	37	13
42	32	5	35	15	38	25
43	31	3	36	3	39	14
44	32	0	36	1	37	27
46	32	1	35	1	37	2
4	31	1	35	1	37	28
10	32	1	35	1	38	74
12	31	1	34	5	37	7
13	33	1	35	1	37	19
18	32	2	35	2	37	2
29	32	1	35	1	37	6
45	32	2	35	1	38	33
47	32	1	35	9	38	89

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:

Case No.	D.O.P.	%
26	39	79
30	38	54
37	41	60

CLUMPING OF FAT CELLS

Case No.	D.O.P.	Grade	D.O.P.	Grade	D.O.P.	Grade
1	31	0	34	0	38	2
2	32	1	35	0	37	0
3	32	0	35	0	38	1
5	32	0	35	0	37	2
6	31	0	35	0	37	0
7	33	0	36	0	38	2
8	31	0	34	0	37	0
9	32	0	35	0	37	1
11	31	0	34	0	37	2
14	32	0	34	0	37	0
15	32	0	35	-	38	1
16	32	0	34	0	37	1
17	33	0	35	0	37	2
19	31	0	35	1	36	2
20	32	0	35	0	37	2
21	32	0	35	0	37	0
22	32	0	35	0	37	0
23	32	0	35	-	37	2
24	33	0	35	0	37	0
25	31	0	34	0	37	0
26	32	0	35	0	37	0
27	32	0	34	0	36	0
28	32	0	36	0	37	1
30	32	0	35	0	37	1
31	31	0	34	0	37	0
32	32	0	35	0	37	1
33	31	0	34	0	-	-
34	31	0	35	0	37	1
35	32	0	35	0	37	1
36	32	0	35	0	37	1
37	31	0	34	0	37	0
38	32	0	35	0	-	-
39	31	0	35	0	37	1
40	32	0	35	0	37	1
41	32	0	35	1	37	1
42	32	0	35	0	38	0
43	31	0	36	0	39	2
44	32	0	36	0	37	1
46	32	0	35	0	37	0
4	31	0	35	0	37	0
10	32	0	35	0	38	2
12	31	0	34	0	37	0
13	33	0	35	0	37	1
18	32	0	35	0	37	0
29	32	0	35	0	37	0
45	32	0	35	0	38	1
47	32	0	35	0	38	2

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:

Case No.	D.O.P.	Grade
26	39	1
30	38	2
37	41	2

POLYGONAL CELLS

Case No.	D.O.P.	Fig. %	D.O.P.	Fig. %	D.O.P.	Fig. %
1	31	0	34	0	38	48
2	32	20	35	22	37	17
3	32	2	35	4	38	2
5	32	0	35	2	37	40
6	31	1	35	3	37	4
7	33	0	36	5	38	7
8	31	0	34	2	37	5
9	32	0	35	1	37	1
11	31	0	34	0	37	0
14	32	1	34	0	37	0
15	32	0	35	-	38	22
16	32	0	34	1	37	21
17	33	0	35	1	37	55
19	31	1	35	4	36	10
20	32	1	35	3	37	20
21	32	1	35	1	37	5
22	32	2	35	3	37	6
23	32	0	35	-	37	5
24	33	2	35	5	37	20
25	31	1	34	1	37	27
26	32	1	35	1	37	5
27	32	1	34	-	36	3
28	32	1	36	5	37	9
30	32	5	35	7	37	24
31	31	1	34	2	37	-
32	32	5	35	5	37	16
33	31	2	34	7	-	-
34	31	4	35	12	37	54
35	32	2	35	3	37	12
36	32	8	35	13	37	7
37	31	0	34	3	37	4
38	32	1	35	18	-	-
39	31	0	35	3	37	2
40	32	0	35	1	37	10
41	32	1	35	6	37	7
42	32	11	35	9	38	15
43	31	1	36	5	39	61
44	32	1	36	1	37	23
46	32	1	35	1	37	5
4	31	0	35	0	37	1
10	32	0	35	0	38	40
12	31	0	34	6	37	4
13	33	0	35	2	37	8
18	32	1	35	2	37	1
29	32	0	35	1	37	2
45	32	1	35	2	38	27
47	32	1	35	8	38	76

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:

Case No.	D.O.P.	Fig. %
26	39	62
30	38	67
37	41	69

ANUCLEATE SQUAMES

Case No.	D.O.P.	Fig. %	D.O.P.	Fig. %	D.O.P.	Fig. %
1	31	24	34	36	38	39
2	32	24	35	24	37	15
3	32	13	35	16	38	25
5	32	15	35	25	37	12
6	31	21	35	21	37	16
7	33	26	36	24	38	21
8	31	18	34	25	37	24
9	32	19	35	23	37	27
11	31	20	34	17	37	29
14	32	16	34	22	37	24
15	32	27	35	-	38	21
16	32	34	34	28	37	20
17	33	22	35	23	37	10
19	31	34	35	24	36	21
20	32	16	35	26	37	30
21	32	34	35	26	37	32
22	32	33	35	29	37	23
23	32	27	35	-	37	24
24	33	37	35	27	37	21
25	31	35	34	19	37	26
26	32	26	35	30	37	32
27	32	21	34	-	36	16
28	32	21	36	21	37	21
30	32	19	35	17	37	21
31	31	26	34	26	37	-
32	32	13	35	11	37	16
33	31	21	34	26	-	-
34	31	16	35	21	37	8
35	32	21	35	18	37	12
36	32	16	35	23	37	18
37	31	29	34	23	37	23
38	32	20	35	28	-	-
39	31	17	35	19	37	19
40	32	40	35	19	37	13
41	32	18	35	13	37	12
42	32	20	35	6	38	14
43	31	25	36	21	39	12
44	32	27	36	22	37	13
46	32	12	35	19	37	16
4	31	32	35	29	37	31
10	32	18	35	11	38	11
12	31	38	34	39	37	35
13	33	28	35	26	37	33
18	32	19	35	24	37	20
29	32	20	35	24	37	22
45	32	15	35	15	38	12
47	32	19	35	21	38	11

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:

Case No.	D.O.P.	Fig. %
26	39	16
30	38	8
37	41	4

SUPERFICIAL SQUAMES

Case No.	D.O.P.	Fig. %	D.O.P.	Fig. %	D.O.P.	Fig. %
1	31	37	34	33	38	35
2	32	20	35	15	37	23
3	32	40	35	40	38	46
5	32	34	35	28	37	19
6	31	38	35	45	37	41
7	33	37	36	34	38	32
8	31	46	34	25	37	29
9	32	49	35	40	37	40
11	31	44	34	48	37	37
14	32	38	34	35	37	30
15	32	33	35	-	38	15
16	32	31	34	34	37	24
17	33	41	35	42	37	17
19	31	40	35	31	36	29
20	32	37	35	31	37	27
21	32	40	35	52	37	53
22	32	46	35	33	37	39
23	32	33	35	-	37	43
24	33	46	35	29	37	30
25	31	36	34	46	37	23
26	32	38	35	42	37	45
27	32	42	34	-	36	41
28	32	51	36	41	37	35
30	32	40	35	33	37	27
31	31	32	34	41	37	-
32	32	38	35	41	37	35
33	31	38	34	37	-	-
34	31	53	35	36	37	17
35	32	34	35	31	37	27
36	32	31	35	26	37	32
37	31	34	34	34	37	28
38	32	35	35	25	-	-
39	31	40	35	33	37	34
40	32	27	35	33	37	31
41	32	37	35	31	37	32
42	32	27	35	28	38	28
43	31	30	36	41	39	11
44	32	26	36	21	37	22
46	32	28	35	26	37	24
4	31	39	35	29	37	32
10	32	33	35	26	38	16
12	31	25	34	15	37	26
13	33	29	35	30	37	21
18	32	33	35	39	37	41
29	32	51	35	53	37	53
45	32	40	35	41	38	28
47	32	28	35	31	38	3

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:

Case No.	D.O.P.	Fig. %
26	39	15
30	38	13
37	41	11

INTERMEDIATE SQUAMES

Case No.	D.O.P.	Fig. %	D.O.P.	Fig. %	D.O.P.	Fig. %
1	31	38	34	29	38	24
2	32	34	35	38	37	44
3	32	43	35	38	38	26
5	32	49	35	40	37	28
6	31	37	35	27	37	37
7	33	31	36	36	38	38
8	31	34	34	47	37	40
9	32	31	35	33	37	30
11	31	33	34	33	37	33
14	32	41	34	42	37	45
15	32	37	35	-	38	41
16	32	34	34	36	37	33
17	33	30	35	31	37	17
19	31	24	35	40	36	38
20	32	38	35	37	37	20
21	32	24	35	18	37	8
22	32	17	35	33	37	30
23	32	37	35	-	37	27
24	33	15	35	38	37	29
25	31	27	34	31	37	21
26	32	32	35	25	37	17
27	32	34	34	-	36	40
28	32	25	36	32	37	33
30	32	33	35	40	37	27
31	31	40	34	30	37	-
32	32	41	35	42	37	32
33	31	38	34	28	-	-
34	31	24	35	28	37	20
35	32	42	35	47	37	47
36	32	39	35	36	37	42
37	31	36	34	39	37	43
38	32	41	35	28	-	-
39	31	42	35	43	37	44
40	32	32	35	46	37	45
41	32	42	35	48	37	48
42	32	41	35	56	38	42
43	31	43	36	32	39	15
44	32	46	36	56	37	41
46	32	58	35	54	37	54
4	31	27	35	40	37	36
10	32	46	35	61	38	32
12	31	35	34	38	37	33
13	33	40	35	39	37	36
18	32	46	35	34	37	36
29	32	27	35	20	37	22
45	32	41	35	39	38	30
47	32	50	35	39	38	10

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:	Case No.	D.O.P.	Fig. %
	26	39	7
	30	38	11
	37	41	15

BASAL CELLS

Case No.	D.O.P.	Fig. %	D.O.P.	Fig. %	D.O.P.	Fig. %
1	31	1	34	1	38	1
2	32	2	35	1	37	1
3	32	1	35	2	38	1
5	32	5	35	5	37	1
6	31	2	35	3	37	2
7	33	5	36	1	38	2
8	31	1	34	1	37	2
9	32	1	35	3	37	1
11	31	3	34	2	37	1
14	32	4	34	1	37	1
15	32	2	35	-	38	1
16	32	1	34	1	37	2
17	33	7	35	3	37	1
19	31	1	35	1	36	2
20	32	8	35	3	37	3
21	32	1	35	3	37	2
22	32	2	35	2	37	2
23	32	2	35	-	37	1
24	33	0	35	1	37	0
25	31	1	34	3	37	2
26	32	3	35	2	37	1
27	32	2	34	-	36	0
28	32	2	36	1	37	2
30	32	3	35	3	37	1
31	31	1	34	1	37	-
32	32	3	35	1	37	1
33	31	1	34	2	-	-
34	31	3	35	3	37	1
35	32	1	35	1	37	2
36	32	6	35	2	37	1
37	31	1	34	1	37	2
38	32	3	35	1	-	-
39	31	1	35	2	37	1
40	32	1	35	1	37	1
41	32	2	35	2	37	1
42	32	1	35	1	38	1
43	31	1	36	1	39	1
44	32	0	36	0	37	1
46	32	1	35	0	37	1
4	31	2	35	1	37	0
10	32	3	35	2	38	1
12	31	2	34	2	37	2
13	33	2	35	2	37	2
18	32	1	35	1	37	2
29	32	2	35	2	37	1
45	32	3	35	3	38	2
47	32	2	35	1	38	0

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:

Case No.	D.O.P.	Fig. %
26	39	0
30	38	1
37	41	1

LIQUOR UREA

Case No.	D.O.P.	mg%	D.O.P.	mg%	D.O.P.	mg%
1	31	32.8	34	34.4	38	29.5
2	32	28.4	35	37.4	37	42.0
3	32	20.4	35	35.8	38	54.5
5	32	23.6	35	40.0	37	30.4
6	31	27.2	35	22.2	37	32.6
7	33	28.8	36	41.0	38	30.0
8	31	19.9	34	14.4	37	34.9
9	32	25.7	35	31.0	37	32.0
11	31	23.3	34	28.0	37	39.0
14	32	36.0	34	20.4	37	32.0
15	32	18.4	35	23.8	38	31.1
16	32	21.4	34	17.7	37	28.2
17	33	30.2	35	35.2	37	33.7
19	31	25.6	35	35.0	36	22.9
20	32	31.4	35	30.8	37	31.9
21	32	19.2	35	23.3	37	36.3
22	32	32.2	35	31.2	37	30.8
23	32	32.0	35	29.5	37	33.4
24	33	41.8	35	41.0	37	48.5
25	31	26.2	34	27.0	37	48.5
26	32	31.5	35	26.3	37	36.8
27	32	20.3	34	25.2	36	40.0
28	32	39.5	36	36.9	37	51.4
30	32	25.5	35	26.0	37	41.9
31	31	32.8	34	27.9	37	35.5
32	32	24.7	35	25.8	37	36.8
33	31	24.6	34	24.5	-	-
34	31	24.6	35	34.9	37	31.6
35	32	35.4	35	36.0	37	37.6
36	32	31.6	35	32.5	37	33.1
37	31	26.1	34	20.2	37	33.3
38	32	31.0	35	35.2	-	-
39	31	29.0	35	31.4	37	35.7
40	32	31.0	35	36.4	37	43.2
41	32	32.6	35	28.7	37	25.5
42	32	24.9	35	24.2	38	31.8
43	31	29.3	36	27.6	39	35.8
44	32	30.3	36	31.1	37	31.8
46	32	37.1	35	32.9	37	35.6
4	31	26.8	35	22.9	37	24.4
10	32	37.2	35	35.8	38	66.2
12	31	28.8	34	41.0	37	26.5
13	33	36.4	35	28.2	37	43.8
18	32	30.1	35	47.8	37	46.4
29	32	29.6	35	33.2	37	34.5
45	32	23.6	35	24.5	38	35.5
47	32	38.4	35	33.1	38	47.0

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:

Case No.	D.O.P.	mg%
26	39	35.0
30	38	46.1
37	41	29.5

MATERNAL UREA

Case No.	D.O.P.	mg%	D.O.P.	mg%	D.O.P.	mg%
1	31	14.0	34	16.7	38	13.8
2	32	14.3	35	18.8	37	23.6
3	32	16.2	35	23.0	38	23.8
5	32	13.4	35	38.2	37	16.2
6	31	20.6	35	12.8	37	23.0
7	33	20.6	36	22.6	38	15.1
8	31	16.8	34	8.8	37	10.5
9	32	16.2	35	24.0	37	14.3
11	31	16.8	34	19.6	37	19.7
14	32	22.2	34	14.9	37	18.2
15	32	16.5	35	15.5	38	22.4
16	32	17.1	34	10.2	37	20.0
17	33	18.0	35	18.4	37	18.6
19	31	20.4	35	19.6	36	15.3
20	32	16.4	35	19.6	37	21.9
21	32	15.2	35	15.2	37	17.6
22	32	25.3	35	23.3	37	13.9
23	32	21.2	35	19.3	37	18.6
24	33	23.7	35	23.2	37	29.0
25	31	17.5	34	16.5	37	22.7
26	32	16.2	35	14.5	37	24.1
27	32	18.2	34	22.9	36	24.4
28	32	23.8	36	24.8	37	35.4
30	32	17.5	35	23.1	37	15.5
31	31	23.4	34	21.5	37	19.2
32	32	20.6	35	21.3	37	19.7
33	31	17.6	34	18.0	-	-
34	31	16.7	35	22.7	37	15.5
35	32	26.3	35	21.7	37	20.8
36	32	20.6	35	18.2	37	18.5
37	31	17.4	34	10.7	37	15.2
38	32	29.8	35	25.8	-	-
39	31	22.8	35	22.6	37	21.4
40	32	23.4	35	25.4	37	21.8
41	32	27.2	35	18.0	37	21.5
42	32	14.9	35	15.9	38	24.5
43	31	19.0	36	14.3	39	22.7
44	32	18.1	36	17.2	37	18.1
46	32	25.2	35	20.9	37	16.8
4	31	23.4	35	17.8	37	13.0
10	32	24.4	35	19.3	38	40.0
12	31	15.3	34	22.4	37	18.3
13	33	22.7	35	23.0	37	26.9
18	32	18.6	35	22.2	37	24.5
29	32	18.4	35	24.2	37	26.3
45	32	22.3	35	20.5	38	28.2
47	32	31.0	35	21.5	38	29.0

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:	Case No.	D.O.P.	mg%
	26	39	29.3
	30	38	16.4
	37	41	18.8

LIQUOR : MATERNAL UREA DIFFERENCE

Case No.	D.O.P.	mg%	D.O.P.	mg%	D.O.P.	mg%
1	31	18.8	34	17.7	38	15.7
2	32	14.1	35	18.6	37	18.4
3	32	4.2	35	12.8	38	30.7
5	32	10.2	35	11.8	37	14.2
6	31	6.6	35	9.4	37	9.4
7	33	8.2	36	18.4	38	14.9
8	31	3.1	34	5.6	37	24.4
9	32	9.5	35	7.0	37	17.7
11	31	6.5	34	8.4	37	19.3
14	32	13.8	34	5.5	37	13.8
15	32	1.9	35	8.3	38	8.7
16	32	4.3	34	7.5	37	8.2
17	33	12.2	35	16.8	37	15.1
19	31	5.2	35	15.4	36	7.6
20	32	15.0	35	11.2	37	10.0
21	32	4.0	35	8.1	37	18.7
22	32	6.9	35	7.9	37	16.9
23	32	10.8	35	9.9	37	14.8
24	33	18.1	35	17.8	37	19.5
25	31	8.7	34	10.5	37	25.8
26	32	15.3	35	11.8	37	12.7
27	32	2.0	34	2.3	36	15.6
28	32	15.7	36	12.1	37	16.0
30	32	8.0	35	2.9	37	26.4
31	31	9.4	34	6.4	37	16.3
32	32	4.1	35	4.5	37	17.1
33	31	7.0	34	5.5	-	-
34	31	7.9	35	12.2	37	16.1
35	32	9.1	35	14.3	37	16.8
36	32	11.0	35	14.3	37	14.6
37	31	8.7	34	9.5	37	18.1
38	32	1.2	35	9.4	-	-
39	31	6.2	35	8.8	37	14.3
40	32	7.6	35	11.0	37	21.4
41	32	5.4	35	10.7	37	4.0
42	32	10.0	35	8.3	38	7.3
43	31	10.3	36	13.3	39	13.1
44	32	12.2	36	13.9	37	13.7
46	32	11.9	35	12.0	37	18.8
4	31	3.4	35	5.1	37	11.4
10	32	12.8	35	16.5	38	26.2
12	31	13.5	34	18.6	37	8.2
13	33	13.7	35	5.2	37	16.9
18	32	11.5	35	25.6	37	21.9
29	32	11.2	35	9.0	37	8.2
45	32	1.3	35	4.0	38	7.3
47	32	7.4	35	12.0	38	18.0

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:

Case No.	D.O.P.	mg%
26	39	6.7
30	38	19.7
37	41	10.7

LIQUOR CREATININE

Case No.	D.O.P.	Fig. mg%	D.O.P.	Fig. mg%	D.O.P.	Fig. mg%
1	31	1.22	34	1.09	38	1.51
2	32	0.43	35	2.08	37	3.10
3	32	1.22	35	1.83	38	2.93
5	32	1.54	35	2.24	37	3.44
6	31	1.10	35	1.44	37	1.82
7	33	1.74	36	1.59	38	1.38
8	31	0.93	34	0.99	37	1.13
9	32	1.36	35	1.61	37	1.68
11	31	1.55	34	1.65	37	2.60
14	32	1.30	34	1.41	37	2.04
15	32	1.21	35	2.09	38	2.48
16	32	1.04	34	1.67	37	1.73
17	33	1.37	35	1.34	37	1.52
19	31	0.42	35	1.94	36	2.00
20	32	1.14	35	1.98	37	1.76
21	32	1.37	35	1.71	37	2.33
22	32	1.62	35	1.64	37	1.96
23	32	1.35	35	2.02	37	2.19
24	33	1.68	35	2.02	37	2.52
25	31	1.60	34	1.80	37	2.57
26	32	1.70	35	1.90	37	2.92
27	32	1.36	34	1.93	36	2.07
28	32	1.68	36	2.13	37	2.44
30	32	1.69	35	2.05	37	2.80
31	31	1.26	34	1.51	37	2.03
32	32	1.62	35	1.90	37	2.12
33	31	1.66	34	2.13	-	-
34	31	1.32	35	1.86	37	2.52
35	32	1.68	35	2.03	37	1.98
36	32	1.77	35	2.09	37	2.28
37	31	0.83	34	1.32	37	2.18
38	32	1.74	35	1.88	-	-
39	31	1.62	35	2.24	37	2.06
40	32	1.17	35	1.96	37	2.70
41	32	1.54	35	1.50	37	2.08
42	32	1.26	35	1.89	38	2.00
43	31	1.52	36	1.39	39	2.40
44	32	2.00	36	1.72	37	2.18
46	32	1.70	35	1.95	37	2.08
4	31	1.01	35	1.59	37	2.24
10	32	1.17	35	1.84	38	2.08
12	31	1.16	34	1.54	37	1.92
13	33	1.78	35	1.70	37	1.94
18	32	1.56	35	1.95	37	2.56
29	32	1.56	35	1.58	37	1.84
45	32	0.94	35	1.62	38	2.20
47	32	1.60	35	1.58	38	1.50

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:

Case No.	D.O.P.	Fig. mg%
26	39	2.32
30	38	3.03
37	41	2.06

MATERNAL TRUE CREATININE

Case No.	D.O.P.	Fig. mg%	D.O.P.	Fig. mg%	D.O.P.	Fig. mg%
1	31	0.55	34	0.53	38	0.20
2	32	0.69	35	0.57	37	0.64
3	32	0.53	35	1.00	38	0.73
5	32	0.58	35	0.72	37	0.65
6	31	0.48	35	0.37	37	0.56
7	33	0.48	36	0.42	38	0.64
8	31	0.48	34	0.41	37	0.49
9	32	0.46	35	0.57	37	0.45
11	31	0.62	34	0.59	37	0.80
14	32	0.43	34	0.48	37	0.69
15	32	0.56	35	0.73	38	0.58
16	32	0.51	34	0.57	37	0.49
17	33	0.57	35	0.53	37	0.52
19	31	0.52	35	0.66	36	0.54
20	32	0.38	35	0.42	37	0.38
21	32	0.65	35	0.47	37	0.68
22	32	0.56	35	0.64	37	0.67
23	32	0.45	35	0.48	37	0.36
24	33	0.72	35	0.68	37	0.84
25	31	0.60	34	0.59	37	0.48
26	32	0.78	35	0.75	37	0.91
27	32	0.48	34	0.61	36	0.63
28	32	0.74	36	0.81	37	0.79
30	32	0.83	35	0.60	37	0.90
31	31	0.55	34	0.51	37	0.65
32	32	0.62	35	0.94	37	0.73
33	31	0.39	34	0.61	-	-
34	31	0.56	35	0.52	37	0.34
35	32	0.68	35	0.84	37	0.88
36	32	0.66	35	0.73	37	0.84
37	31	0.40	34	0.52	37	0.70
38	32	0.88	35	1.04	-	-
39	31	0.54	35	0.76	37	0.72
40	32	0.69	35	0.68	37	0.70
41	32	0.65	35	0.52	37	0.72
42	32	0.58	35	0.57	38	0.60
43	31	0.55	36	0.57	39	0.64
44	32	0.60	36	0.67	37	0.80
46	32	0.68	35	0.70	37	0.74
4	31	0.54	35	0.20	37	0.63
10	32	0.50	35	0.68	38	0.84
12	31	0.51	34	0.65	37	0.43
13	33	1.00	35	0.41	37	0.74
18	32	0.25	35	0.52	37	0.56
29	32	0.65	35	0.59	37	0.55
45	32	0.58	35	0.55	38	0.51
47	32	0.87	35	0.53	38	0.80

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:

Case No.	D.O.P.	Fig. mg%
26	39	1.00
30	38	0.81
37	41	0.66

LIQUOR TRUE CREATININE

Case No.	D.O.P.	Fig. mg%	D.O.P.	Fig. mg%	D.O.P.	Fig. mg%
1	31	0.89	34	1.05	38	1.33
2	32	1.33	35	2.01	37	2.43
3	32	1.15	35	1.77	38	2.73
5	32	1.29	35	2.20	37	2.28
6	31	0.97	35	1.20	37	1.56
7	33	1.12	36	1.26	38	1.94
8	31	0.93	34	1.26	37	1.51
9	32	1.15	35	1.46	37	1.51
11	31	1.14	34	1.33	37	1.74
14	32	1.17	34	1.40	37	1.68
15	32	0.86	35	1.53	38	2.12
16	32	0.96	34	1.14	37	1.24
17	33	1.42	35	1.54	37	1.75
19	31	1.40	35	1.64	36	2.00
20	32	0.79	35	1.16	37	1.23
21	32	0.93	35	1.31	37	2.08
22	32	1.10	35	1.61	37	1.95
23	32	1.10	35	1.58	37	1.65
24	33	1.08	35	1.42	37	1.73
25	31	0.88	34	1.65	37	1.68
26	32	1.34	35	1.98	37	2.00
27	32	0.98	34	1.11	36	2.00
28	32	1.40	36	1.85	37	2.58
30	32	1.30	35	1.91	37	2.19
31	31	0.85	34	1.50	37	1.53
32	32	1.09	35	1.45	37	1.89
33	31	1.12	34	2.00	-	-
34	31	1.18	35	1.57	37	1.69
35	32	1.19	35	1.55	37	1.87
36	32	1.26	35	1.59	37	1.77
37	31	0.74	34	1.10	37	1.40
38	32	1.27	35	1.68	-	-
39	31	1.05	35	1.43	37	1.76
40	32	1.17	35	1.54	37	1.87
41	32	1.07	35	1.45	37	2.26
42	32	0.94	35	1.33	38	1.68
43	31	1.08	36	1.32	39	1.69
44	32	1.18	36	1.66	37	1.77
46	32	1.40	35	1.71	37	1.98
4	31	0.88	35	1.37	37	1.61
10	32	1.35	35	1.50	38	1.53
12	31	0.95	34	1.04	37	1.31
13	33	1.44	35	1.46	37	1.66
18	32	0.67	35	1.07	37	1.76
29	32	1.24	35	1.25	37	1.66
45	32	0.76	35	1.28	38	1.60
47	32	1.04	35	1.32	38	1.90

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:	Case No.	D.O.P.	Fig. mg%
	26	39	2.46
	30	38	2.37
	37	41	1.79

LIQUOR : MATERNAL TRUE CREATININE DIFFERENCE

Case No.	D.O.P.	Fig. mg%	D.O.P.	Fig. mg%	D.O.P.	Fig. mg%
1	31	0.34	34	0.52	38	1.13
2	32	0.64	35	1.44	37	1.79
3	32	0.62	35	0.77	38	2.00
5	32	0.71	35	1.48	37	1.63
6	31	0.49	35	0.83	37	1.00
7	33	0.64	36	0.84	38	1.30
8	31	0.45	34	0.85	37	1.02
9	32	0.69	35	0.89	37	1.06
11	31	0.52	34	0.74	37	0.94
14	32	0.74	34	0.92	37	0.99
15	32	0.30	35	0.80	38	1.54
16	32	0.45	34	0.57	37	0.75
17	33	0.85	35	1.01	37	1.23
19	31	0.88	35	0.98	36	1.46
20	32	0.41	35	0.74	37	0.85
21	32	0.28	35	0.84	37	1.40
22	32	0.54	35	0.97	37	1.28
23	32	0.65	35	1.10	37	1.29
24	33	0.36	35	0.74	37	0.89
25	31	0.28	34	1.06	37	1.20
26	32	0.56	35	1.23	37	1.09
27	32	0.50	34	0.50	36	1.37
28	32	0.66	36	1.04	37	1.79
30	32	0.47	35	1.31	37	1.29
31	31	0.30	34	0.99	37	0.88
32	32	0.47	36	0.51	37	1.16
33	31	0.73	34	1.39	-	-
34	31	0.62	35	1.05	37	1.35
35	32	0.51	35	0.71	37	0.99
36	32	0.60	35	0.86	37	0.93
37	31	0.34	34	0.58	37	0.70
38	32	0.39	35	0.64	-	-
39	31	0.51	35	0.67	37	1.04
40	32	0.48	35	0.86	37	1.17
41	32	0.42	35	0.93	37	1.54
42	32	0.36	35	0.76	38	1.08
43	31	0.53	36	0.75	39	1.05
44	32	0.58	36	0.99	37	0.97
46	32	0.79	35	1.01	37	1.24
4	31	0.34	35	1.17	37	0.98
10	32	0.85	35	0.82	38	0.69
12	31	0.44	34	0.39	37	0.88
13	33	0.44	35	1.05	37	0.92
18	32	0.42	35	0.55	37	1.20
29	32	0.59	35	0.66	37	1.11
45	32	0.18	35	0.73	38	1.09
47	32	0.17	35	0.79	38	1.10

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:	Case No.	D.O.P.	Fig. mg%
	26	39	1.46
	30	38	1.56
	37	41	1.13

LIQUOR BILIRUBIN

Case No.	D.O.P.	mg%	D.O.P.	mg%	D.O.P.	mg%
1	31	0.04	34	0.02	38	0.01
2	32	0.08	35	0.13	37	0.20
3	32	0.09	35	0.06	38	0.04
5	32	0.15	35	0.19	37	0.08
6	31	0.01	35	0.03	37	0.02
7	33	0.05	36	0.01	38	0.08
8	31	0.07	34	0.09	37	0.02
9	32	0.08	35	0.13	37	0.22
11	31	0.09	34	0.05	37	0.33
14	32	0.04	34	0.02	37	0.02
15	32	0.01	35	0.02	38	0.02
16	32	0.08	34	0.02	37	0.14
17	33	0.05	35	0.04	37	0.09
19	31	0.11	35	0.04	36	0.02
20	32	0.01	35	0.05	37	0.05
21	32	0.01	35	0.08	37	0.08
22	32	0.08	35	0.05	37	0.02
23	32	0.02	35	0.18	37	0.15
24	33	0.01	35	0.02	37	0.02
25	31	0.16	34	0.02	37	0.01
26	32	0.04	35	0.07	37	0.02
27	32	0.07	34	0.04	36	0.07
28	32	0.05	36	0.08	37	0.02
30	32	0.03	35	0.02	37	0.08
31	31	0.03	34	0.08	37	0.02
32	32	0.03	35	0.02	37	0.08
33	31	0.03	34	0.03	-	-
34	31	0.21	35	0.17	37	0.07
35	32	0.02	35	0.02	37	0.01
36	32	0.13	35	0.10	37	0.03
37	31	0.11	34	0.02	37	0.01
38	32	0.02	35	0.02	-	-
39	31	0.03	35	0.05	37	0.01
40	32	0.10	35	0.01	37	0.03
41	32	0.01	35	0.02	37	0.02
42	32	0.05	35	0.02	38	0.01
43	31	0.18	36	0.10	39	0.08
44	32	0.10	36	0.03	37	0.02
46	32	0.02	35	0.18	37	0.08
4	31	0.04	35	0.06	37	0.10
10	32	0.01	35	0.01	38	0.01
12	31	0.02	34	0.05	37	0.12
13	33	0.05	35	0.02	37	0.08
18	32	0.08	35	0.18	37	0.02
29	32	0.05	35	0.02	37	0.03
45	32	0.07	35	0.27	38	0.08
47	32	0.13	35	0.08	38	0.08

Case Nos. 26, 30 and 37 had a fourth result as explained on page 9.

These were:

Case No.	D.O.P.	mg%
26	39	0.07
30	38	0.04
37	41	0.02

TABLE XX

MEANS AND STANDARD DEVIATIONS OF VARIABLES MEASURED AT 30-33 WEEKS; 34-36 WEEKS AND 37+ WEEKS FOR NORMAL PATIENTS

	30 - 33 Weeks		34 - 36 Weeks		37+ Weeks	
	Mean	S.D.	Mean	S.D.	Mean	S.D.
H.O.F.	31.7	1.0	34.7	1.0	36.8	1.0
State of cervix	1.0	0.2	1.2	0.4	1.6	0.8
Fat globules	0.2	0.4	0.3	0.5	1.2	1.0
Blue squames	97.4	4.6	94.2	6.5	80.0	17.0
Yellow anucleates	0.2	0.6	0.1	0.4	0.8	2.4
Fat cells	2.3	4.7	5.7	6.4	19.2	15.6
Clumping of fat cells	0.0	0.2	0.1	0.2	0.9	0.7
Polygonal cells	2.1	3.8	4.6	4.9	17.0	16.8
Anucleate cells	22.8	7.2	21.8	5.7	19.6	7.3
Superficial cells	36.7	6.9	34.7	8.0	30.5	10.0
Intermediate cells	36.1	8.0	37.2	8.8	33.0	11.3
Basal cells	2.1	1.8	1.7	1.1	1.3	0.6
Amnion cells	0.2	0.4	0.1	0.2	0.1	0.3
Maternal urea	19.4	3.9	19.2	4.6	20.0	4.7
Maternal true creatinine	0.58	0.12	0.63	0.16	0.65	0.17
Maternal bilirubin	0.28	0.15	0.31	0.20	0.33	0.2
Liquor urea	28.3	5.6	30.2	6.7	36.0	6.7
Liquor creatinine	1.39	0.35	1.79	0.31	2.23	0.50
Liquor true creatinine	1.11	0.19	1.54	0.28	1.82	0.35
Liquor bilirubin	0.06	0.05	0.06	0.05	0.06	0.07
Ultrasonic B.P.D.	87.3	2.9	93.5	3.6	96.7	2.9
Uterine volume	4028	749	5001	746	5746	820

TABLE XXI

MEANS AND STANDARD DEVIATIONS OF VARIABLES MEASURED AT 30-33 WEEKS; 34-36 WEEKS AND 37+ WEEKS FOR ABNORMAL PATIENTS

	30 - 33 Weeks		34 - 36 Weeks		37+ Weeks	
	Mean	S.D.	Mean	S.D.	Mean	S.D.
H.O.F.	30.8	0.9	33.8	0.8	35.4	0.7
State of cervix	1.0	0.0	1.1	0.3	1.8	0.7
Fat globules	00.1	0.3	0.1	0.4	0.7	1.0
Blue squames	98.6	0.5	97.8	1.8	69.4	31.7
Yellow anucleates	0.0	0.0	0.1	0.4	1.2	3.0
Fat cells	1.3	0.5	2.1	1.8	28.3	29.6
Clumping of fat cells	0.0	0.0	0.0	0.0	0.6	0.7
Polygonal cells	0.9	0.8	2.0	1.9	18.3	25.5
Anucleate cells	23.3	8.5	24.6	8.7	21.9	9.6
Superficial cells	35.6	9.2	33.3	11.3	28.8	14.8
Intermediate cells	38.1	12.4	38.0	11.3	29.4	8.5
Basal cells	2.1	0.8	1.9	0.6	1.4	0.9
Amnion cells	0.0	0.0	0.3	0.5	0.1	0.3
Maternal urea	22.4	4.6	21.5	2.2	24.5	8.4
Maternal true creatinine	0.61	0.22	0.53	0.16	0.64	0.14
Maternal bilirubin	0.19	0.11	0.25	0.09	0.29	0.13
Liquor urea	31.5	5.02	33.1	8.4	39.5	13.0
Liquor creatinine	1.38	0.31	1.68	0.14	2.03	0.30
Liquor true creatinine	1.05	0.26	1.32	0.20	1.66	0.19
Liquor bilirubin	0.06	0.04	0.08	0.09	0.06	0.04
Ultrasonic B.P.D.	82.0	4.4	87.3	3.8	92.3	2.9
Uterine volume	3183	698	3968	722	4396	807

APPENDIX F

A preliminary study into the accuracy of the menstrual histories of patients delivery in the Peninsula Maternity Services hospital group.

A premature baby was for a long time defined, by international agreement, as one weighing 2,500 grams or less at birth regardless of the length of gestation. It has become apparent, however, that this group of babies can be divided into the truly premature or short gestation, and the dysmature or 'light-for-dates' infants. This division has a very practical use as the two sorts of baby behave very differently, and are liable to different complications in the immediate post-natal period.

Since the introduction by Farr et al (1966) and Dubowitz et al (1970) of accurate methods of assessment of gestational age in the newborn infant, the paediatrician can be statistically forewarned of the problems an individual small baby is likely to manifest. This is achieved by ageing the baby after delivery and classifying it into one of the abovementioned categories.

The accuracy of this paediatric assessment of gestational age prompted the author to use it as a means of verifying the menstrual histories of a sample of patients delivered in the above unit.

Materials and Methods

All neonates born in our unit are now gestationally aged by means of the method described by Dubowitz et al (1970).

Six hundred and thirty-eight randomly selected patient folders were drawn from the records of two of the hospitals in our unit. Two hundred and seventy-six were those of European* patients delivered

at the Mowbray Maternity Hospital (M.M.H.) and 362 were folders of non-European* patients delivered at Groote Schuur Hospital (G.S.H.). There was no selection whatsoever, so that factors such as age, parity, socio-economic status, complications of pregnancy etc. were not assessed.

At her first visit, each patient is asked the date of her last normal menstrual period (L.N.M.P.) and whether she is sure or unsure of the date thereof. Retrospectively the calculated gestational age at delivery was noted as was the paediatric assessment of gestational age. If a patient delivered within two weeks of the expected date of delivery (E.D.D.) as calculated by Nägele's rule, then her L.N.M.P. was regarded as being correct. Correlation between the calculated gestational age and that derived by paediatric examination of the neonate, was taken as evidence of a correct L.N.M.P. when delivery occurred outside the range 38 - 42 weeks. All other instances were diagnosed as incorrect menstrual histories.

Results

Table I indicates the proportions of patients who were sure or unsure of the date of their L.N.M.P. and those patients who could give no date at all.

TABLE I

The subjective reliability of menstrual histories supplied by the patients at their booking visit

Hospital	Number of patients	Subjective menstrual history reliability		
		Sure	Unsure	Unknown
M.M.H.	276	199 (72.1%)	63 (22.8%)	14 (5.1%)
G.S.H.	362	262 (72.4%)	38 (10.5%)	62 (17.1%)

Table II shows what proportion of each of the above subjective categories was considered to be wrong by two weeks or more. Obviously those patients who could provide no date at all had to be considered to be wrong, as an index of the reliability of menstrual histories was being sought.

TABLE II

The percentage of patients whose dates were found to be incorrect

Hospital	Sure of dates yet wrong	Unsure of dates and wrong	Total of patients with wrong dates
M.M.H.	48 (24.1%)	31 (49.2%)	93 (33.7%)
G.S.H.	87 (33.2%)	18 (47.4%)	167 (46.1%)

Of the European patients who were able to give some date when asked for the L.N.M.P., 30.15% were shown to be wrong. The figure for the non-Europeans was 35.5% of the dates provided.

Conclusion

From this very brief assessment, it would seem that our Europeans tend to give a more reliable menstrual history than our non-Europeans. It would appear that just under half our patients (1,848 Europeans and 13,278 non-Europeans in 1970) are totally unreliable in respect of their menstrual histories.

It is pertinent to note that 41% of these 638 patients paid their first visit to the antenatal clinics after 20 weeks of gestation.

* For the purposes of this study, and the main research project, the Europeans referred to are Caucasoid and the non-Europeans are non-Caucasoid.

APPENDIX G

Measurement of the approximate dose of ionizing radiation received by a fetus from a single abdominal exposure.

In an attempt to discover what sort of dose of ionizing radiation a fetus was exposed to during the taking of a single abdominal exposure for the purposes of assessing its gestational age radiologically, the author persuaded the Biophysics Department to attempt such a measurement.

The X-ray unit at Mowbray Maternity Hospital was used as the experiment would have caused too great a disruption of the already hard-pressed radiological unit at Groote Schuur Hospital.

Thermoluminescent dosemeters (Lithium fluoride powder) were used to measure the dose of X-rays to which a wax and bolus phantom were exposed. To obtain a graph of the results, three hypothetical patients were created; small, medium and large. The focal-film distance (F.F.D.) was maintained at 100cm but the exposure time and dose of X-rays delivered were varied as they would be for three such patients.

Figure 1 shows diagramatically, the measurements for the experiment. It was felt the dose measured in the model should approximate that received by a fetus even allowing for different degrees of penetration.

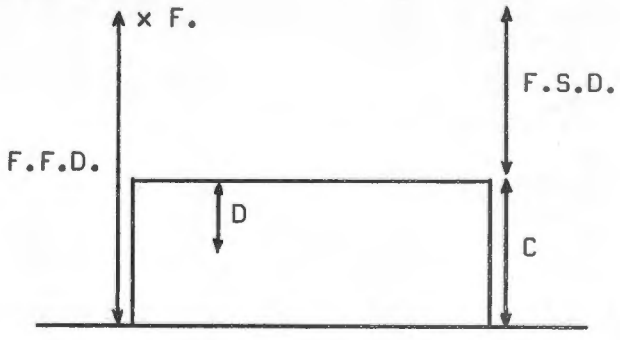


Figure 1 Diagramatic representation of the model.

C : Diameter or 'thickness' of the patient in centimetres

D : Distance from surface to fetus in centimetres

F.F.D. Focal-film distance in centimetres

F.S.D. Distance from anode to subject (F.F.D. - C.) in centimetres

The results of the three experiments are listed below.

A. Small patient - (60mAs., 75kV)

F.S.D. = 80cm

C = 20cm

D = 10cm

Dose at skin surface = 1.30 Rads

Dose to fetus = 0.25 Rads

B. Medium patient - (60mAs., 80kV)

F.S.D. = 72cm

C = 28cm

D = 12cm

Dose at skin surface = 1.60 Rads

Dose to fetus = 0.31 Rads

C. Large patient - (75mAs., 87kV)

F.S.D. = 62cm

C = 38cm

D = 14cm

Dose at skin surface = 3.10 Rads

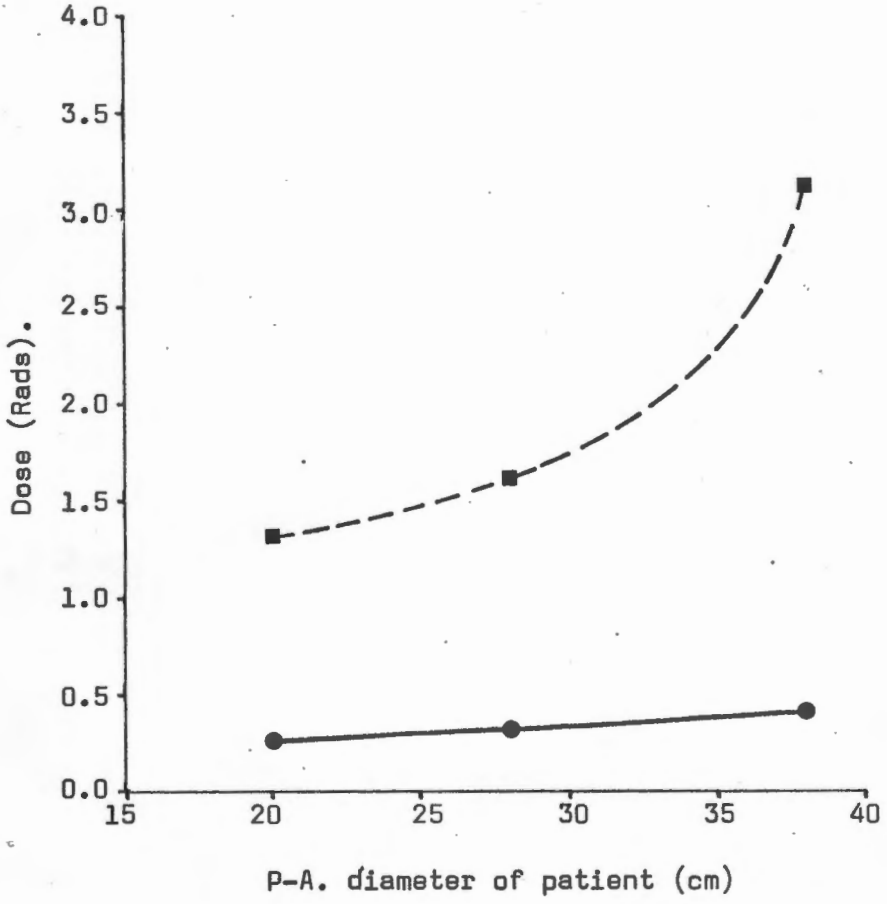
Dose to fetus = 0.40 Rads

These results were then plotted (figure 2) so that some idea of the dose received by the fetus in patients within this range, could be obtained.

It can thus be seen, that when using the P-A view in the radiological assessment of gestational age in subjects within the above experimental range, the dose of ionizing radiation received by the fetus is less than 0.5 Rads. This is obviously doubled if a second exposure is required, for example, when the patient is rotated to throw the fetal knee joints clear of the maternal spine.

The author wishes to thank Mrs. M. de Villiers for conducting this experiment.

Figure 2



Graphic representation of the dose of ionizing radiation received by mothers and fetuses during examination of X-ray maturity.

Dose at skin surface

Dose to fetus