

THE EFFECTS OF GLUTAMIC ACID ON THE INHERITANCE OF
BEHAVIOUR TENDENCIES IN THE WHITE RAT.



by

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

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INTRODUCTION.

The inheritance of learning ability and temperament is a subject of great interest and importance to students of psychology as well as to geneticists.

The genetic basis of behaviour tendencies which constitute temperament provides psychologists with an extremely complex problem as the genetic constitution is diluted considerably by environmental forces. Social standards and conventions differ unsystematically from one community to another, and also within the same community, therefore, the dilution is itself not systematic. Thus, if the psychologist wishes to postulate theories regarding temperament, he is faced with a long and arduous task before a satisfactory theory will result.

For these reasons he turns to humbler animals to formulate his laws and theories. The most popular of these animals is the white rat because of its peculiar advantages for research purposes. "It is clean, easy to house and handle, and it is inexpensive to maintain." (1) Because of its fecundity large numbers from one generation are available for comparative purposes; where we can have well-matched groups with respect to sex, age and genetic constitution. As the gestation period in the rat is only about twenty-one days, studies on the inheritance of

behaviour/ ...

behaviour tendencies, involving several generations can be carried out in a comparatively short time. Donaldson (2) states that the physiological processes in the rat are basically the same as in man, therefore, "similarities between mammals having the same food habits tend to be close, and that in some instances at least, by the use of equivalent ages, the results obtained with one form can be very precisely transferred to the other." These comments are applicable also to the fundamental processes of behaviour, whose nature cannot be solved, either in man or the rat, until the physiological mechanisms have been uncovered.

Because complicated habits are absent in the rat, investigations of many problems are easier than in man, and techniques of observation can reach higher degrees of accuracy. The animal can be treated as an instrument of measurement in stimulus-response relationships and can be controlled as thoroughly as the stimuli themselves. In the rat we can control genetic constitution, destroy neural structures, sense organs and glands, control the whole environment and do whatever is necessary in our attempts to understand the fundamental processes basic to behaviour. We can supplement our knowledge of human subjects by results obtained in the animal field, and thus advance one step further in our understanding of human behaviour ~~behaviour~~ problems.

During the course of experiments carried out in the Psychological Laboratory at the University of Cape Town to obtain data for the analysis of behavioural oscillation, it was noticed that the results showed two distinct groups of behaviour. The differences in the behaviour proved to be qualitative as well as quantitative, for on further investigation, it was seen that these differences could be linked up with emotional responses. Hence, it was concluded that the animals had spontaneously divided themselves into two distinct temperamental types. These two types were designated A and B, to avoid describing the temperamental differences in terms analagous to human temperamental differences. In consequence of the discovery of the two lines or temperaments, it was decided to start a programme of selective breeding to try to develop these lines. The A males were mated with the A females, and the B males with the B females. Their offspring were put in the same experimental situation that produced the observed differences, and the classification of the animals in each generation into A and B types was based on their behaviour in the experimental situation. By carrying on this process of selective breeding it was found that after a sufficient length of time these two lines were purified.

Once we have two pure strains, we know what to expect

of the /...

of the behaviour of the offspring, and thus, we can keep the hereditary factor constant and vary the environmental factor as desired. If the classification of these types is based on their hereditary reactions to particular maze situations, then it follows, as Heron (3) states, that "these two strains must differ in some physiological, neurological, or biochemical respect or respects, or in a combination of all. If the animals are examined by techniques belonging to these various fields, it may be possible to determine the existence of such differences. We may, therefore, be able to determine the somatic variations underlying different degrees of maze learning ability."

Two important factors were maintained in the breeding programme, the females were not required to produce more than one litter, to preserve the virility of the groups, and each female was put with two males in a separate breeding cage; the males were changed once a week.

The differences between A and B types are attributable to the functioning of the nervous system, and if glutamic acid has any effect on the Central Nervous system, we would expect some deviation in the behaviour of the B's.

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CHAPTER I.

THE PROBLEM TO BE INVESTIGATED.

We know that the offspring of the B animals are usually of the B type. The B type of behaviour shows itself in the reaction to a reduced reward, latency goes up steeply to infinity. Also the reaction to handling shows strong emotional components. The problem is this. Glutamic acid is known to have an effect on the maze learning capacity of rats, it increases liveliness in general, and improves performance. The problem is to determine whether this effect can be produced through the mother, that is to say, if glutamic acid is fed to the mothers, is there a significant improvement in the performance of the offspring as compared to that of the control group. If glutamic acid fed to the mothers is absorbed by the young, then there are two possible ways in which this can happen. Either during prenatal life, where the glutamic acid is taken into the bloodstream of the fetus, or secondly, it can be transmitted through the milk, or both. If it is transmitted through the bloodstream during the prenatal life, there should be a significant difference in the offspring of mothers who are fed glutamic acid during pregnancy and those who are given glutamic acid during the lactation period.

Then/ ..

Then, if it is transmitted through the bloodstream, presumably there will be a difference in the offspring of mothers that get glutamic acid before conception, or, for a period before conception, through pregnancy, during lactation, and mothers who get glutamic acid only when the first signs of pregnancy appear.

To test this hypothesis, we have four groups of mothers.

- (1) This first group is given glutamic acid before conceiving right up to the end of lactation.
- (2) Here, glutamic acid is administered after pregnancy and right through to the end of lactation.
- (3) In this group, glutamic acid given only during lactation.
- (4) Which is the Control group, receives no glutamic acid at all.

We must then test the reactions of the offspring to determine whether any change or improvement has occurred, if so, this can be attributed to the glutamic acid.

CHAPTER II.

REVIEW OF THE LITERATURE ON GLUTAMIC ACID.

Glutamic acid has been listed by the nutritionists as a non-essential amino-acid as it is considered not essential for growth. But Womack and Rose (1) have shown that glutamic acid exerts statistically significant effects upon the growth of rats deprived of arginine, one of the essential amino-acids. It, however, is not as effective a growth stimulant as is arginine. In an experiment involving 300 animals and lasting 28 days, the mean growth increment induced by arginine was 29.1 gm., while that for glutamic acid only 8.9 gm. They discovered that arginine, proline (another non-essential amino-acid) and glutamic acid are mutually interconvertible in the organism of the rat, but at different rates as exemplified by their different influence upon growth. The glutamic acid data obtained by them show, for the first time, that this amino-acid may be transformed IN VIVO into proline or arginine. Comparisons of the effects of arginine alone, and of arginine, proline, hydroxyproline, and glutamic acid administered simultaneously, demonstrate that the rate of gain is moderately improved when all four amino-acids are present in the food. The superiority of the latter ration is due largely, if not entirely, to the inclusion of glutamic acid.

Hydroxyproline/

Hydroxyproline has been shown to be a dispensable amino-acid. Proline also belongs to the dispensable group, but, in the growing rat, can replace in part the arginine requirement. The status of glutamic acid is less certain. Tentatively, it is regarded as non-essential, but its final classification must await the results of additional experiment.

Stern et al (2) have stated that glutamic acid appears to be an important substance in cerebral metabolism. Evidence obtained suggests that when the glucose supply fails, glutamic acid can be utilised by the brain, it is the only amino-acid metabolised by the brain.

It was found too, that when glutamic acid was administered to eight patients, they became more attentive, alert, and persistent in trying to accomplish the tasks put before them, emotional control also appeared to improve.

Price, Waelsch and Putnam (3) reported that dl-glutamic acid hydrochloride may be used as an adjuvant in the treatment of petit mal seizures of epileptic patients who are otherwise resistant to anticonvulsant therapy.

Zimmerman and Ross (4) decided to use glutamic acid on rats and to demonstrate the effect of its ingestion on maze learning in the normal rat, the dosage was 200 mg.

Their/

Their experiment consisted of two parts. In the first part they used four groups of animals, the first group was fed dl-pyrrolidine carboxylic acid, containing 29 rats, the second group, was fed neutralised dl-pyrrolidine carboxylic acid, 16 rats, the third group, was fed aminoacetic acid, 20 rats, and the fourth group, which was the control, consisted of 27 rats. The dl-pyrrolidine carboxylic acid is derived from racemic glutamic acid.

The rats fed dl-pyrrolidine carboxylic acid and the rats fed neutralised dl-pyrrolidine carboxylic acid, showed approximately the same order of means and standard deviations. While, the rats fed aminoacetic acid and the control rats showed the same general tendencies. The mean number of trials required to learn the maze was five trials for the rats fed dl-pyrrolidine carboxylic acid, four trials, for those fed neutralised dl-pyrrolidine carboxylic acid, seventeen trials for the rats fed aminoacetic acid and fourteen trials for the control. The difference in the number of trials for the rats fed dl-pyrrolidine carboxylic acid and the number for the rats fed neutralised dl-pyrrolidine carboxylic acid is not significant, and does not, in all probability, represent a real difference.

The results/ ..

The results in terms of time required, the number of trials and the errors were all statistically significant. No difference in learning was found between rats fed neutralised and rats fed unneutralised dl-pyrrolidine carboxylic acid.

The unnatural amino-acids are not metabolised to any great extent in the animal body. It is probable, therefore, that only the natural isomer of pyrrolidine carboxylic acid, the l form, is metabolised and that only this part of the compound administered accounts for the effects observed. This view is supported by the observation that l-pyrrolidine carboxylic acid when fed to rabbits is not excreted in the urine. It is possible that this substance is reconverted into natural glutamic acid. So, they concluded that the effect with l-pyrrolidine carboxylic acid may be due to that of l-glutamic acid.

The second part of their experiment was concerned with studying the effect of ingestion of naturally occurring l-glutamic acid on maze learning in the white rat. They found that there was no difference of statistical significance between the performance of these rats and those that were fed dl-pyrrolidine carboxylic acid.

Encouraged by these results, Zimmerman and Ross (5) proceeded to investigate the effect of glutamic acid on mental functioning in human subjects. They found that
on verbal/

on verbal, motor and personality tests given, definite improvements, following glutamic acid therapy, could be observed in each of the nine subjects during a six month interval. They found an average rise in the Intelligence Quotient of nine points when using the Stanford-Binet Test. The greatest improvement in intelligence occurred during the first six months, after which the acceleration was diminished and appeared to approach its limit after one year of treatment.

Further recent investigations have shown that "the release of acetylcholine is intrinsically connected with the electrical changes during nerve activity, that is, it generates the electric changes occurring everywhere at the neuronal surface." Also, that "acetylcholine is intrinsically connected with nerve activity and that an increased rate of formation of acetylcholine in the presence of glutamic acid has been demonstrated IN VITRO." (3) Therefore, any physiological changes that might occur, may be related to the formation of acetylcholine, which is brought about by the presence of glutamic acid.

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3. /

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CHAPTER III.

DISCUSSION OF A AND B TYPES OF BEHAVIOUR.

The main difference between the A and B types lies in the fact that the B type of behaviour appears to be an emotional response to a frustrating situation, such as one involving a negligible food incentive. Here, reactive inhibition overtakes both effective jumping and jumping reaction potential. They display emotionality too in other situations. When an attempt is made to pick up a B rat, it cowers in a corner, sways from side to side, and once it is caught it bites, squeals, defecates, urinates and wriggles to free itself. Whereas, A animals after an initial disturbance adapt themselves to a frustrating situation. A certain amount of I_R is generated at first, but S_{H_R} soon overtakes and supersedes it. The A's try to evade capture by darting backwards and forwards, but when caught, they do not display emotionality or attempt to escape.

The animals are placed in the experimental situation and the classification of them into A and B types is based on the course of the jumping latency curve. If the curve falls after an initial rise, the animal is classified as an A; while, if it rises continuously, as a linear function of the number of trials, the animal is of the B type.

During the/

During the course of several experiments carried out in the Psychological Laboratory, it was discovered that the B type of behaviour is brought to light only when the reduced reward falls below some limiting value, this value was found to be .6gm.

A further distinguishing feature is that the proportion of A animals that learn the shorter path, is much greater than the proportion of the B animals.

Other differences are given in the following table, taken from Mr. Taylor's Paper. (1).

Situation.	Behaviour.		
	A	B	INTERMEDIATE.
About to be caught.	Darting movements to avoid capture.	Cowering in a corner.	Ineffective escape movements.
In the experimenter's hand.	Calm submission.	Squirming, squealing, defecating, etc.	Sometimes docile, sometimes aggressive.
1st day in the maze.	Lively exploratory movements.	Slow hopping cautious movements, standing at corners and in food-box entrances, with body stretched, head on one side, and swaying.	Irresolute hesitations alternating with jerky but rapid exploratory movements.
1st day on jumping platform.	Lively exploratory movements.	Agitated climbing on walls, and clawing, chattering, defecating, etc.	A and B behaviours alternating unpredictably.

Continued.

Situation.	A	B	<u>INTERMEDIATE.</u>
Some days after beginning of Expt. in cages, when about to be caught.	Marked readiness to be picked up.	Cowering in corners.	Alternation.
Mating.	Quick reactions.	Markedly slower reactions. Conception a day later than the A's.	No record.
Litters.	Range 8 to 13 pups. Average about 10.	Range 5 to 10. Average about 7.	No record.

These differences have been attributed to genetic factors. Taylor (1) suggested that there is a multiple gene mechanism determining the A and B types, the A behaviour is caused by the preponderance of alleles of one type, while B behaviour by a preponderance of alleles of an alternative type. As the A females possess a larger complement of the alleles than the A males, and the B females similarly have a larger complement of the alleles responsible for the B behaviour, the progeny of these types are mainly of the maternal type. The preponderance of males in the progeny of A females, and females in the progeny of B females, he thought was due to the chemical constitution of the genital tract of A females which may

favour/ ...

favour spermatozoa bearing the Y chromosome, while that of the B females favours the X chromosome.

The difference in the size of the litters in the A and B types was attributed to the higher level of neural activity in the A's which would also tend to increase the rate of ovulation.

What is inherited is some general characteristic of the nervous system. In an experiment in which luminal was administered to A rats, it was found that their behaviour was indistinguishable from that of the B animals. Since luminal depresses the central nervous system, this shows that the B animals generate a larger amount of inhibitory potential than the A animals.

As reaction latency is the criterion used for distinguishing the A and B types of behaviour, we shall consider it here very briefly.

The jumping habit is increased by virtue of the reinforcement at each trial, and it is subject to diminution by virtue of the reduced reward. A reduced reward produces frustration which tends to generate reactive inhibition, and this in turn lessens the strength of S_{HR} . In the A animals S_{HR} and S_{ER} become sufficiently strong during the preliminary training to counteract the accumulation of I_R during frustration. Whereas, in the

B animals/

B animals jumping generates large amounts of reactive inhibition, which tends to become conditioned. The reaction potential will be diminished by the amount of inhibition that was conditioned at the previous trial. A certain amount of inhibition is learned at the 2nd trial, this being learned remains. Most of the inhibition generated disappears, but some of it is carried over and acts against the S^E_R which is available for jumping. The sum of these two inhibitions operates against jumping at the 3rd trial, and so on. As there is a definite amount of conditioned inhibition added at each trial, the total conditioned inhibition is, therefore, a linear function of the number of trials.

Therefore, in the case of B animals in a frustration situation, in this case a negligible food incentive, the rate of accumulation of I_R will be greater than the rate of increase of S^H_R , so that S^E_R diminishes with successive trials and falls eventually below the reaction threshold, when the animals will stop jumping.

In the A animals, the accumulation of I_R is more rapid at first than the increase of S^H_R in frustration situations, but S^H_R soon catches up and supersedes I_R .

Dr. Reichlin (2) states that if S^E_R is insufficient to mediate a jump at a given moment, it may yet be enough to mediate preparatory movement, so that at a low level of S^E_R many such movements are likely to be made before

the animal/..

the animal finally jumps. When the element of choice is involved, deciding here between alternative paths, these movements take the form of Vicarious Trial and Error movements. These movements are in effect preparatory reactions leading to a jump, but involve less expenditure of energy than the act of jumping. Therefore, the same S^E_R that mediates jumping, mediates the preparatory movements also. Since these preparatory movements involve less expenditure of energy, the reaction threshold for a VTE movement will be lower than that for jumping. Thus when only a small proportion of the S^O_R distribution curve is above the jumping threshold, the animal might not jump, but it will VTE.

In the B animals when S^E_R falls as shown by increased latencies, and S^O_R distribution falls below the S^L_R , VTE movements will still occur, but once it falls below the VTE threshold, these responses will cease, and the animal will remain huddled and inactive on the jumping platform.

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CHAPTER IV.

THE RESEARCH PROGRAMME.

The purpose of the experiment was to determine what effects l-glutamic acid would have on the offspring of B mothers fed glutamic at three different stages. Namely, before conception, two weeks after pregnancy, and finally at lactation.

Nine females were selected, seven being of the pure B strain while the other two were diluted B's, since their parentage was mixed, they were from A mothers and B fathers. These two females were used as the Control. All the females were mated with pure B males. A record was made of the weights at different stages during the experiment.

Zimmerman has shown that l-glutamic acid produced the same results as did dl-pyrrolidine carboxylic acid, so we decided to use the former in our experiment. The dosage was 300 mg. The tablets were powdered and made into small pellets with some moistened mash, these pellets were placed on the bare floor of separate feeding boxes to ensure that each female ate all her ration. The pregnant mothers ate their pellets readily, while the pre-conception females had to be coaxed by hand. They were greatly distressed and disturbed, and displayed all the emotional tendencies, defecating, urinating, squirming and biting. By the eighth day, however, they were more resigned/ ..

resigned to handling and started to eat their pellets without encouragement.

At the first signs of pregnancy, the females were separated from the males and were placed in separate cages. During the last week of pregnancy they were left in their living cages to avoid injury. A section was cleared in the living cages for the pellets and the experimenter watched the females every day to see that they consumed the pellets. These were always given an hour before the ordinary ration for the day. This ration consisted of 6 grams of mash for every 30 grams of body weight.

Included in the experimental animals was one pregnant intermediate female, she was given glutamic acid at two weeks pregnancy. This was just to compare the behaviour of her offspring with that of the others.

Group I. (Pre-conception group.) The 3 females in this group were given glutamic acid three days before mating, starting on the 17th April, and finishing at the end of lactation on the 11th June.

Group II. (Two weeks pregnant.) This group consisted of 2 experimental females together with the extra female mentioned above. Glutamic acid was administered on the 17th April for the first time, and was continued until 16th May when lactation ended.

Group III/ ...

Group III. (Lactation). These two females were given glutamic acid on the 27th April, when their pups were born, and ended on the 18th May, when they were weaned.

The lactation period in all groups was approximately three weeks, and if the rats were not weaned naturally by that time, they were separated from their mothers. The baby rats were not at any time given glutamic acid.

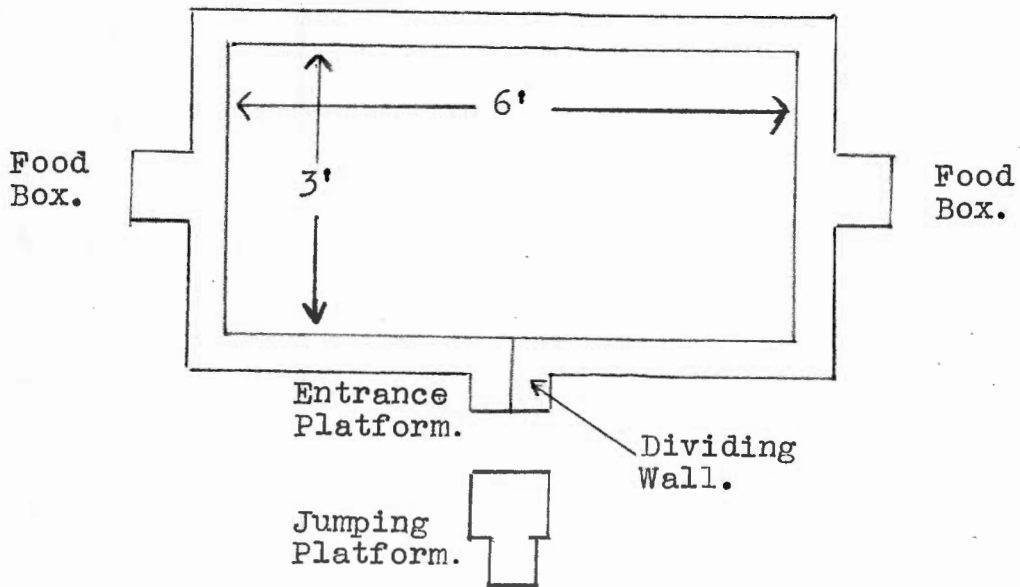
The offspring were given the normal ration, which consisted of mash for the first week, and then five hard biscuits each, together with one piece of carrot and half a cabbage leaf, daily, until the period of preliminary training.

This commenced once the rats were six weeks old. They were all given exactly the same preliminary training.

The maze used was the simple rectangular one, with food-boxes at each side, equidistant from the entrance. The animal enters the maze from a jumping platform which can be set at any distance from the maze. A soft net was hung between the platform and the maze to protect the animals from injury or shock in the event of a fall. In this way a fall could not be inhibitory to future jumps and thus fear of jumping the gap was avoided. A dividing wall on the entrance platform separates the left-hand alley from the right, so that after the animal has jumped only one path is open to it.

The dimensions/ ..

The dimensions of the maze are shown in the diagram below.



During the preliminary training the dividing wall was removed and four grams of food were put in each box. The training was designed to induce the rats to jump over an 8 inch gap from platform to maze. At first the gap was only 1 inch wide, so that the rats could walk across it, and then increased by one inch every day until it reached 8 inches. This training period lasted for 14 days, at the end of which all the animals were jumping within three seconds after being placed on the jumping platform.

For the first three days the animals were trained in groups to prevent fear and to aid adjustment to a new situation which could perhaps arouse obstructive emotionality.

After this/

After this they were all marked and run in the same order every day. Recordings were made of any direction bias during the preliminary training, by the end of which preference for any direction was firmly established.

From the beginning of training the animals were given half rations per gram body weight.

In the experiment proper, with the dividing wall, which lasted for 14 days, 0.4 gm. of food was placed in one box only, and in the opposite direction to the direction bias shown in the preliminary training. So that when a rat showed preference for jumping right, without the dividing wall, the food was placed in the left box and vice versa. The distance of 8 inches was held constant throughout the experiment. The measurements recorded included jumping latency, the path chosen and the time spent in the maze before reaching the box containing food.

Latency, was measured in the following way. The rat was placed at the rear of the jumping platform, which is walled in on three sides, and as soon as it entered the front compartment of the maze, the stop-watch, recording tenths of a second, was started, and it was stopped when the animal jumped. Another stop-watch was used for recording the time spent in the maze, this was started when the rat landed on the entrance, and was stopped when it entered the food-box, the time spent in the maze was also recorded in

tenths/

tenths of a second. The path chosen was recorded as "short" or "long" if the rat jumped to the left when the food was in the left-hand box, this was recorded as the short path, but, if it jumped to the right, then it had chosen the long path.

Unfortunately, the numbers of the offspring for each group were not the same, they are as follows.

Group I (Pre-conception) - three females.

(a) Had ten pups, one was diminutive being half the size of the others in the litter; this pup (its sex unknown) was eaten by the mother. There were 4 males and 5 females in this litter.

(b) Had a litter of five pups, consisting of 3 males and 2 females. They were larger than the offspring of (a) and (c).

(c) Had a litter of seven pups, consisting of 2 males and 5 females. All strong, but not as large as the (b) litter.

TOTAL number of rats used for the experiment in the preconception group was 21.

Because of the smallness of the numbers in all the groups, males and females were used. The female's oestrus cycle begins at the end of the third month by which time the animals had finished the experiment, and therefore, it could not have had any effect on their behaviour. As no statistically significant differences in the learning ability of the sexes have been established, both males and females were used confidently.

Group II (Two weeks pregnant) consisted of two females -

(a) This one had a litter of 8 pups, with 3 males and 5 females.

(b) /

- (b) This litter consisted of 7 pups, with 6 males and only 1 female.

The pups in both these groups were well-formed and healthy.

Included in this group was an Intermediate female, who had 14 pups, and this broke the Department record. This large number proved too difficult for her to manage, so she threw three pups out of the nest, and although the experimenter repeatedly put them back, she allowed them to die. As they were very young, their sex could not be determined. The remainder consisted of 6 males and 5 females.

TOTAL number of experimental rats in this group was 15, together with the extra 11 rats, whose recordings and behaviour were treated as another group. Hereafter called Group IIB.

Group III(Lactation). This group consisted of two females whose offspring were as follows.

- (a) Three pups in this litter, being 1 male and 2 females.
- (b) Had a litter of 5 pups, with 4 males and 1 female.

TOTAL number of experimental rats in this group was 8.

Group IV (Control). There were two females in this group.

- (a) Had four pups, 2 males and 2 females.
- (b) Had 3 pups, 2 males and 1 female. These pups were much larger than those in any other group.

TOTAL number of rats in experimentag control group was 7.

During/ ;

During the breeding programme in the Psychological laboratory it was found that the number of pups in the litters of B animals, ranged from five to ten, with an average of seven. In this experiment, the average for the first two groups is seven, and that for the third group, four. Thus glutamic acid does not appear to have affected the reproductive organs. But, the intermediate female fed glutamic acid at two weeks' pregnancy had fourteen pups, the average for A animals is ten, while no average is known for the intermediate animals, therefore, this increase might be attributable to increased ovulation due to the glutamic acid.

TO SECURE OPTIMAL EXPERIMENTAL CONDITIONS THE FOLLOWING FACTORS WERE CONSIDERED.

1. HANDLING.

The animals were weaned at three weeks, and were handled daily by the experimenter for the next three weeks to accustom them to this aspect of the situation, and thus to obviate any handling factor. No other persons were permitted to handle the animals.

2. FEEDING.

The feeding times were kept regular, and during the preliminary training only half the required amount per gram body weight was given. The hunger period was kept constant at 23 hours. The range of motivation most facilitative to learning lies between 1 and 24 hours privation

of food/..

of food.

3. AGE.

All the animals were six weeks old at the beginning of the preliminary training, and eight weeks old when the experiment itself was started. This is regarded as the most suitable age, as the female's oestrus cycle has not yet started, and the period between six weeks and four months is recognised as the period of maximum activity. The younger rats too are free from conflicting habits which arise in older rats, and this tends to make them superior in learning situations.

4. LIVING CONDITIONS.

The living cages were large enough to provide sufficient freedom of movement and plenty of opportunity for exercise. This prevented bursts of reactive activity when placed in the maze situation. They were housed in a laboratory far removed from the general activity in the building, in a room with a constant temperature of about 75°, to prevent any differences in activity or drive due to changes in temperature. The lighting in the room was fluorescent to avoid any shadows and thus exteroceptive stimuli.

5. PRELIMINARY TRAINING.

This was kept constant for all groups, with the animals run in the same order every day. They were allowed to traverse and explore the maze in order to become familiar with it/ ...

with it. Any orientation fixation was avoided as the rats were lifted out of the maze at different points each day.

Their daily ration, which was half ration from the beginning of training, was administered one hour after the training. Each rat had only one trial per day.

The animals were weighed at the beginning of training, and then at the beginning of every week, these weights were kept constant.

They were motivated only by hunger as throughout training and experimentation they were given water. Mash was placed in the food-boxes in the maze to prevent delay caused by the necessity to drink water while eating hard food. Their daily ration consisted of rat biscuit, containing vitamins, minerals and all the necessary proteins, together with fresh cabbage and diced carrots. In this way their health was maintained in a satisfactory condition.

6. RECORDINGS.

(a) The jumping latency was recorded as described previously, together with behaviour on the jumping platform, including the number of VTE movements.

(b) The path chosen, whether short or long.

(c) The time in the maze was taken, that is the period the rat took to find the food and enter the food-box, and the behaviour in the maze was also noted.

The recordings/ ..

The recordings on the stop-watch were kept soundless so as not to disturb the animals or to provide any cues.

All conditions were thus controlled to ensure accurate results, there might have been forces operative at the time of which the experimenter was unaware and which could affect the results, but this she feels is most unlikely.

CHAPTER V.

RESULTS.

With regard to the general behaviour glutamic acid does appear to have affected this in some measure.

When handling B animals, they display numerous signs of emotionality, by squirming, wriggling, urinating and defecating, but the animals in this experiment showed these tendencies only during the first four or five days of experimentation. After this, they became less emotional and their behaviour resembled that of the A's.

When placed on the jumping platform they showed consternation by chattering, squealing and defecating. VTE movements were pronounced during the early stages of the experiment, and in some rats, they continued throughout. It appears that some change has occurred since all the rats continued to jump, and with a decrease in the latencies. This suggests that less reactive inhibition was generated and so S^H_R continued to increase. The act of jumping, however, still showed signs of emotionality, for the jumps were sprawled and jerky. In two animals from Group II the jumps were centrally directed, one jumped onto the dividing wall while the other jumped onto the wire mesh above the entrance platform, both forms had disappeared by the tenth trial. The choice of the long path

continued/

continued with few exceptions in all the groups. Both habits were equally reinforced during the preliminary training, since the rats established a direction bias during that time, this particular response was continually reinforced, and the habit of jumping in that direction became strengthened. The shock of finding the preferred box empty does not appear to extinguish the habit of jumping in a particular direction as the negligible reward itself is not strong enough to cause extinction of the long path choice.

In the maze, behaviour of the experimental animals was more akin to the A type than to the B, for they explored it thoroughly, darting about at a high pace, but this activity was goal-directed. There was no cowering, timid or fearful behaviour. They were not distressed by the empty box and soon passed it on their way to the food-box without pausing for inspection.

These facts seem to suggest that glutamic acid had made the rats more active and alert, together with this there appears to be a decrease in emotionality, which is probably due to some alternation in the functioning of the nervous system.

When we consult the tables of results and the average latency curves in Figure I, we see that in all the experimental groups, the latencies tend to decrease.

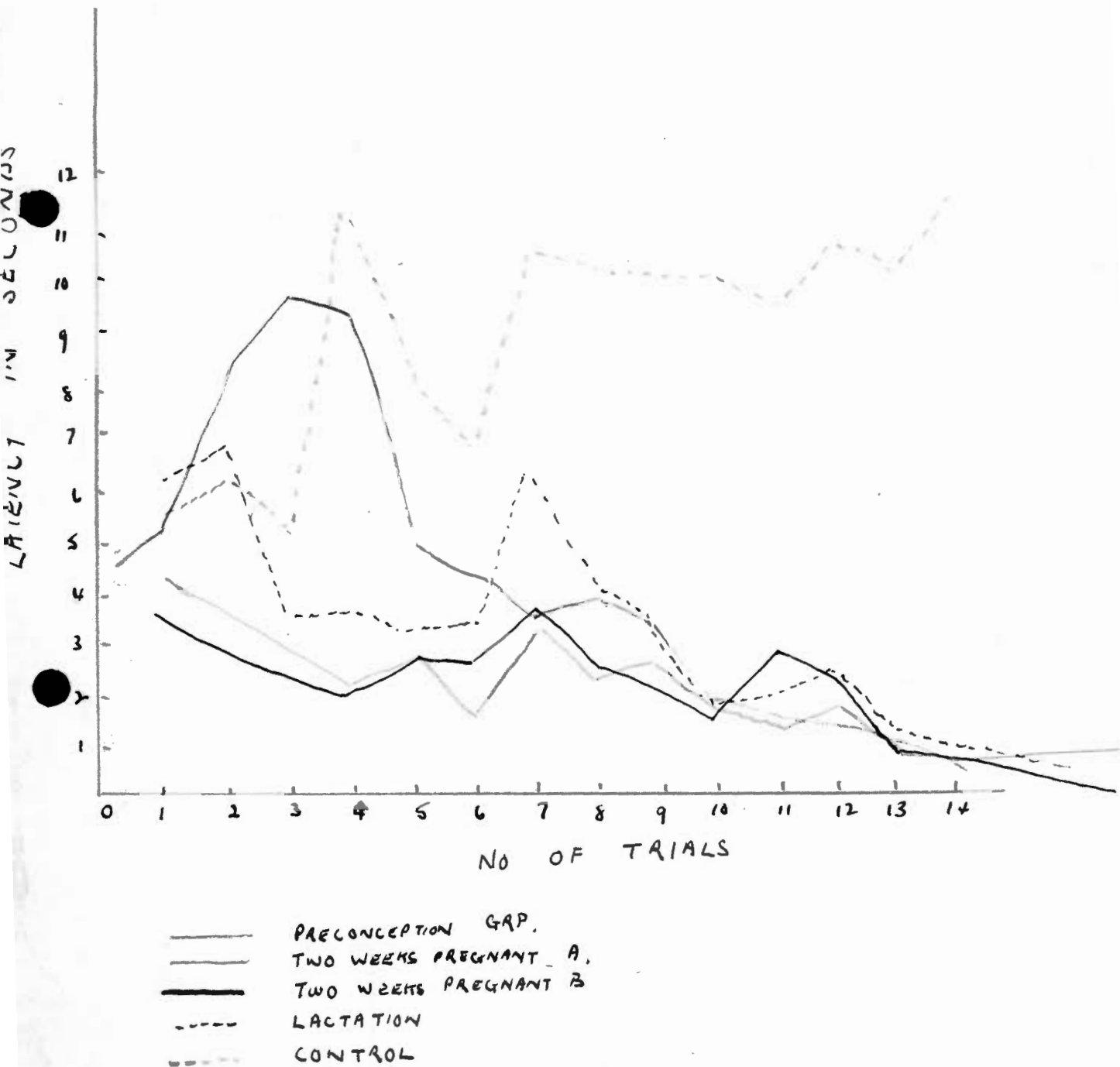


FIGURE I

GROUP I (PRE-CONCEPTION).

Number of animals 21.

TRIALS	LATENCY		SHORT PATH	T. I. M.	
	σ	A		σ	A
1	4.301	5.286	1	6.606	12.542
2	7.694	8.396	2	9.237	12.713
3	5.124	9.610	2	4.778	11.576
4	7.568	9.066	2	4.714	12.367
5	3.412	4.762	1	3.774	7.678
6	2.543	4.362	1	2.719	6.576
7	2.292	3.386	1	1.695	5.607
8	2.742	3.805	1	2.085	6.610
9	2.257	3.138	1	2.139	5.731
10	1.501	1.765	1	1.780	4.537
11	1.185	1.431	2	1.498	4.771
12	.2946	1.224	2	1.380	4.537
13	.5260	.895	1	1.727	4.667
14	.1743	.3095	1	1.231	3.612

GROUP IIA (TWO WEEKS PREGNANT).

Number of Animals. 15

TRIALS	LATENCY.		SHORT PATH.	TIM.	
	σ	A		σ	A.
1	1.699	4.033	3	4.434	10.433
2	2.154	3.400	2	3.454	8.167
3	1.426	2.717	3	2.917	6.300
4	1.508	2.250	3	1.602	4.900
5	1.643	2.750	3	2.526	5.467
6	1.237	1.267	4	3.110	5.046
7	2.062	3.167	3	3.141	5.500
8	1.281	2.150	3	2.449	5.067
9	1.562	2.350	4	2.445	4.300
10	1.348	1.617	4	2.029	4.667
11	.952	1.173	3	1.806	4.667
12	1.143	1.471 \times	3	2.038	4.707
13	.289	.6286	4	2.281	4.214
14	.3824	.5429	3	1.637	3.900

\times From the twelfth day there were only 14 animals in the experiment as one of them cut its throat on the wire water basket.

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GROUP IIb

(INTERMEDIATE - TWO WEEKS PREGNANT.)

Number of Animals 11.

TRIALS	LATENCY.		SHORT PATH.	T. I. M.	
	σ	A		σ	A
1	1.162	3.523	4	4.734	11.636
2	1.841	2.946	3	4.122	8.091
3	.9393	2.541	3	3.450	5.890
4	1.068	2.187	4	1.564	4.591
5	1.189	2.613	5	4.210	5.410
6	1.930	2.614	6	2.021	3.591
7	1.850	3.682	6	4.622	5.410
8	1.972	2.455	6	3.026	4.355
9	1.459	2.141	7	2.740	3.764
10	1.014	1.395	6	2.594	3.400
11	3.742	2.700	4	2.076	4.159
12	2.712	2.073	5	1.070	3.359
13	.2913	.600	5	1.832	3.259
14	.2391	.5091	5	2.071	3.923

GROUP III (LACTATION).

Number of animals 8.

TRIALS	LATENCY		SHORT PATH	T. I. M.	
	σ	A		σ	A
1	3.018	6.025	-	3.666	10.25
2	4.054	6.950	-	2.739	10.50
3	.966	3.438	-	2.088	6.975
4	1.550	3.688	-	3.316	8.000
5	2.595	3.125	1	6.320	9.750
6	2.224	3.375	-	.998	6.063
7	3.999	6.438	1	2.500	5.800
8	2.204	4.375	2	1.557	4.425
9	1.298	3.063	2	1.677	4.450
10	1.670	1.840	1	3.269	5.550
11	1.312	1.900	2	1.031	4.488
12	1.341	2.225	2	2.342	5.525
13	.8874	1.100	2	2.990	5.390
14	.4528	.700	2	1.854	3.850

GROUP IV.
CONTROL.

Number of animals

TRIALS	LATENCY.		7.	T.I.M.	
	σ	A	SHORT PATH.	σ	A
1	2.711	5.465	Nil	4.334	9.714
2	6.093	6.029	"	4.998	12.643
3	2.828	5.000	"	4.998	8.357
4	12.165	11.030	"	5.178	7.871
5	6.312	7.943	"	2.268	6.000
6	6.324	6.800	"	1.359	4.964
7	9.810	10.071	"	1.647	7.429
8	9.486	10.00	"	.961	5.179
9	9.270	9.929	"	1.098	5.036
10	9.270	9.929	"	1.221	4.836
11	9.270	9.129	"	.776	5.179
12	8.637	10.50	"	1.116	5.179
13	9.270	9.929	"	1.134	5.150
14	4.390	11.186	"	1.125	5.207

The curves for Group IIa and Group IIb are almost identical, but the slope of the curves is gradual, suggesting that the rate of building up the habit is slow because a large amount of reactive inhibition is still generated.

In the Pre-conception results, we see that the latency curve drops quickly. Since S^E_R is proportional to the number of reinforcements, the habit of jumping, and doing so quickly, continues to increase because of the almost total absence of reactive inhibition.

The curve for the lactation group is extremely shallow, the latencies do decrease, but not beyond a certain limit. Amounts a reactive inhibition still seem operative here and S^H_R and S^E_R have not become sufficiently strong to combat this.

In the control group, the latency curve rises proportionally to the number of reinforcements. If the experiment had continued for a longer period, this curve probably would have gone on rising. Hence, we see that these animals are generating larger amounts of reactive inhibition than any of the others.

The standard deviations for time in the maze, average time in the maze, which is inversely proportional to S^H_R , all diminish with successive reinforcements.

COMMENTS.

This experiment can be considered as a parallel to the one conducted in the psychological laboratory in which luminal was used, this had a depressing effect on the nervous system, whereas glutamic acid appears to have had a stimulating effect, in that it lessens the amount of inhibitory potential generated in a frustrating situation and thereby increases activity. Probably, it has altered the distribution of the alleles of the B type which has changed the general characteristics of the nervous system.

The change in the nervous system appears to be greater in those animals who received glutamic acid through the bloodstream from mothers who were fed it before conception, less in those whose mothers received it during pregnancy, and even still less in the offspring of the lactation mothers

The sample used was too small to make definite statements, but it does seem possible that glutamic acid could affect the chemical composition of the genital tract which would favour spermatozoa bearing Y chromosomes and thus alter the sex ratio in the B animals.

CONCLUSIONS.

Some improvement has definitely occurred, but since only a small proportion of the rats choose the short path, we can conclude that this improvement in performance

is not/ ..

is not attributable to an increase in the learning ability, but rather to a higher level of activity which is brought about by the effect of glutamic acid on the functioning of the nervous system. This results in the crease of I_R in a situation causing frustration, such as the negligible food reward.
