URINARY HISTAMINE AND GASTRIC SECRETION.

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C O N T E N T S.

LIST OF ILLUSTRATIONS.

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PREFACE.

The work reported in this thesis was carried out over a 2 year period.

The first year was spent in Dr. Code's Laboratory in the Department of Physiology of the Mayo Clinic. I am deeply grateful to Dr. Code for introducing me to this field. His frequent laboratory consultations and the stimulation of his data conferences remain an abiding inspiration to me. I am also indebted to Dr. George Hallenbeck for many stimulating suggestions, and to Dr. Updike whose patient initial studies provided the spring-board for this present work. To Mr. Joe Kennedy I owe a special debt for much help and the gentle way he introduced me to the ways of the Rochester laboratory.

The facilities for my second year's work in Glasgow I owe to Professor Illingworth. In particular, I have to be deeply grateful to him for allowing me to continue these rather unsurgical activities when he had so many surgical research projects requiring workers, and his criticisms and guidance have helped me greatly. I have also to thank Miss May Alexander for her constant help with the tedious task of extracting the histamine from the urine./

/urine.

For their help in preparing the final manuscript, I have to thank Miss Hamilton, Mr. Duthie and the Photographic Department of the Department of Surgery.

CHAPTER I.

INTRODUCTION - HISTAMINE AND GASTRIC SECRETION.

From the vast literature on the physiology of gastric secretion, three definite stimulating pathways have emerged, each playing a part in the acid response of the gastric mucosa to ingestion of a meal.

The psychic secretion of acid produced by sight, smell, taste and swallowing with its efferent stimulation through the vagus was clearly demonstrated by the clinical experiments of Pavlov. Much early evidence had suggested the existence of a humoral or blood-borne stimulus and this "chemical" phase of acid secretion was later separated into gastric and intestinal components.

THE GASTRIC PHASE OF ACID SECRETION.

Edkins, in 1905, discovered that acid and peptone extracts of the pyloric mucosa when injected intravenously produced acid secretion while injection of the extracting substances themselves into the blood stream, did not. He gave the name Gastrin to the active principle involved. The final proof of such a mehcanism was supplied by Ivy and Farrell, 1925, who demonstrated that a subcutaneously auto transplanted pouch of the fundic portion of the stomach secreted hydrochloric acid in response to the presence of /

/of a meal in the main stomach. To determine whether this humoral mechanism was hormonal or whether it was due to absorption of secretagogues from food proved to be a difficult problem because the chemical agents in food which stimulate acid secretion when placed in the stomach are also effective, although less so, when injected intravenously. Crucial physiological proof for the hormonal nature of this humoral mechanism was established by the demonstration that distension of the main stomach, a stimulation which obviously could not be absorbed, could cause the transplanted stomach to secrete (Grossman, Robertson and Ivy, 1948). However, the chemical identity of Gastrin has not yet been determined nor do we know the chemical nature of any of the substances in food which cause significant stimulation of gastric secretion by releasing gastrin. Its site of formation has been considered to be mainly in the pylorus since pylorectomy markedly reduces the response of the total stomach pouch to distension and secretagogues (Grossman and Ivy, 1948).

THE INTESTINAL PHASE OF ACID SECRETION.

It has long been known that many substances introduced directly into the small intestine produce acid secretion. (Babkin, 1928: Ivy, Lim and McCarthy, 1925: Webster and Armour, 1932). Most observers noted a long latent period of 1-3 hours between the beginning of the feed and the start of the acid /

/acid response. Whether this humoral phase is hormonal or due to the absorption of secretagogues has not been clarified. The chemical nature of this intestinal gastrin is not known.

In summary then, one neural and two humoral pathways (the exact chemistry of which are unknown) have been discovered which stimulate acid secretion. Whether these mechanisms produce this effect by acting directly on the acid secreting cells or through release of some intermediary substance is not known.

Since histamine, administered subcutaneously or intravenously has long been known to stimulate acid secretion, many workers have looked for evidence that it might play some part in one or other of these three stimulating mechanisms outlined above. The facts suggesting a possible relationship between histamine and acid secretion will now be reviewed.

HISTAMINE STIMULATES GASTRIC ACID SECRETION.

In 1920 Popielski discovered that histamine stimulated the secretion of acid by the stomach of dogs and this was confirmed by Best and McHenry in 1931. It was later found that the dose response curve relating the rate of injection of histamine to the rate of secretion of gastric juice has the customary exponential character which upon probit transformation becomes a straight line (Obrink, K.J., 1948). /

/ (Obrink, K.J., 1948).

THE SITE OF ACTION OF ADMINISTERED HISTAMINE.

Histamine was found to stimulate gastric mucosa transplanted to subcutaneous tissue (Ivy and Farrell, 1925: Klein, 1932). It even stimulates gastric mucosa, in vitro, in high concentration (Davies, 1946: Davenport and Chavré, 1950). Thus the evidence suggests that histamine acts directly upon the parietal cells, without a chemical transmitter. However, the histamine stimulates the cells much more readily when brought in contact with their submucosal surface, than when applied to their mucosal surface. Thus minute amounts applied to the submucosal surface of the cells stimulate acid production (Hanson, Grossman and Ivy, 1948.)

In contrast tremendous quantities of histamine (50 mg. histamine acid phosphate in 20 ml.) must be applied to the mucosal surface to produce a minimal acid response (Lim, Ivy and McCarthy, 1925).

In summary then histamine stimulates acid secretion, apparently by acting directly on the secretory cells. This has been found to be true for a very wide range of species. According to Code (1955) exceptions are elasmobranch fishes and lower amphibia. Rats and mice, however, respond only to large doses of histamine.

Furthermore, the effect of histamine on gastric /

/gastric secretion appears to be almost limited to stimulation of acid, not pepsin. Thus histamine-induced gastric juice is high in acid and low in pepsin. The acid secretion may wash out some pepsin initially but this increase does not recur if a second injection of histamine is given (Babkin, 1930: Vineberg and Babkin, 1931: Gilman and Cowgill, 1931: Bjorkman, Norden and Uvnas, 1943).

Is there any evidence to suggest that histamine plays a role in the physiological secretion of acid by the gastric mucosa?

HISTAMINE IS PRESENT IN HIGH CONCENTRATION IN THE GASTRIC MUCOSA.

Not only is histamine present in the gastric mucosa, but the acid secreting area (fundus) has twice the concentration of the antrum, which does not secrete acid (Gavin, McHenry and Wilson, 1933: Emmelin and Kahlson, 1944: Trach, Code and Wangenstein, 1944). Recently Feldberg and Harris (1953) have tried to localise the mucosal histamine of the dog's gastro-intestinal tract more exactly by placing flattened pieces of mucosa on the freezing microtome and then cutting the frozen pieces in the horizontal plane in serial sections. The frozen sections were either weighed, extracted and assayed for histamine or stained and examined histologically. From the results of such "histamine profiles" of the gastric mucosa, the peak or highest concentration in the fundic mucosa /

/mucosa appeared in the region of the parietal cells.

It must be noted, however, that these observers found almost as great a concentration in the pyloric mucosa, this time in the region of the non-acid secreting pyloric glands.

HISTAMINASE IS ABSENT FROM THE GASTRIC MUCOSA.

Histaminase is the enzyme which deaminates histamine thus destroying its pharmacological effects (Best, 1939: Best and McHenry, 1930: McHenry and Gavin, 1932). Attempts to demonstrate the presence of significant quantities of histaminase in any part of the gastric mucosa have so far proved unsuccessful. (Best and McHenry, 1930: Rose, Karady and Browne, 1940: Dworetzky and Code, 1951: Waton, 1955).

This would allow minute amounts of histamine, released at the parietal cell or reaching it in the blood stream, every opportunity to stimulate the parietal cell.

DRUGS WHICH INHIBIT HISTAMINASE AUGMENT ACID SECRETION.

In 1947, Grandjean reported that thiamin, Vitamin B_1, potentiated histamine contractions in the guinea pig's ileum. Zeller (1939) had previously observed that thiamin is an inhibitor of histaminase. It remained for Schild to relate these two observations and he and his colleagues elucidated the potentiation of the pharmacological effects of histamine by histaminase inhibitors (Mongar and Schild, 1951: Arunlakshana, /

/Arunlakshana, Mongar and Schild, 1954). The effect of drugs inhibiting histaminase activity, on acid secretion, has been tested. Sircus (1953), using B_1-pyrimidine in dogs and cats, found the response to histamine, meals, vagal stimulation and alcohol were all augmented. Ivy and others, 1956, obtained similar results with aminoguanidine, in dogs.

HISTAMINE IS PRESENT IN GASTRIC JUICE WHATEVER THE STIMULUS.

Histamine has long been recognised in gastric juice (Komarov, 1933: Brown and Smith, 1935: McIntosh, 1938: Emmelin and Kahlson, 1944: Code, Hallenbeck and Gregory, 1947: Adam et al, 1954). Emmelin and Kahlson found the histamine content of the juice to be independent of the mode of stimulus employed in exciting the parietal cells. It was of the same order in juice from the cephalic and gastric phases and in juice obtained by injection of secretagogue drugs. In addition, the histamine concentration of the juice secreted during the cephalic and gastric phases was sufficient to stimulate the parietal cells. Not only does gastric juice contain histamine but there is a definite correlation between secretory activity of the parietal cell and the histaminic activity of the gastric juice (Code, 1955).

To explain these phenomena three different concepts of the possible role of histamine in acid secretion have been developed. /

/developed.

(1) HISTAMINE HAS BEEN CONSIDERED TO BE IDENTICAL WITH GASTRIN.

The fact that histamine was the sole gastric secretory excitant in dilute acid extracts of the pyloric mucosa was taken to indicate that histamine might be identical with "gastrin" by Sachs, Ivy, Burgess and Van Doloh, 1932). They noticed that the vasopressor and secretory properties of their gastrin extract paralleled each other. As stressed by these authors, the crucial question to be answered in this connection is whether the histamine concentration of the blood plasma rises during the gastric phase.

McIntosh, in 1938, using the method of Barsoum and Gaddum, found that the histamine concentration of the systemic blood was not significantly affected by the digestion of a meal but he pointed out that the histamine content of the plasma may have increased without its detection in his experiments where whole blood was used, which gives values representing mainly the histamine content of the corpuscles. However, in experiments in humans, Adam, Card, Riddell, Roberts and Strong failed to find an increase in the histamine content of the plasma, even when extracts were concentrated three and five-fold. McIntosh's findings were confirmed by Emmelin, Kahlson and Wicksell, 1941. These authors gave dogs and cats intravenous infusions of histamine which moderately stimulated acid secretion without any rise in the blood histamine being detected. Adam, 1950, /

/ Adam, 1950, gave histamine subcutaneously and intravenously to humans in doses sufficient to produce headache, increased pulse rate, flushing of the face and a fall in blood pressure without being able to discern any change in the histamine content of the venous blood. In short, the parietal cell is so responsive to histamine, a rise in plasma histamine large enough to produce the secretory response to an ordinary meal might occur without being detected by currently available methods, and these negative blood findings are therefore not conclusive. In passing it should be said that no agreement exists as to whether histamine exists in the blood stream in a free, physiologically active, or in a bound, inactive form. This matter will be discussed in Chapter VIII.

The concept of histamine as a blood borne agent active in the gastric phase has been further weakened by those workers who have isolated a gastrin preparation from the pyloric mucosa, a protein-like substance, apparently free from histamine which, on intravenous injection, causes a profound secretion of acid from the fundic glands (Komarov, 1942: Harper, A.A., 1946: Munch-Petersen, Ronnow and Uvnas, 1944: Jorpes, Jalling and Mutt, 1952).

Further proof that "gastrin" preparations did not owe their secretory activity to traces of histamine was supplied by incubating the gastrin with histaminase before use (Bauer and Uvnas, 1944).

These authors found that quantities of /

/of histaminase which do not affect the secretory power of a "gastrin" preparation destroy an amount of histamine of corresponding secretory activity.

Lastly, quantitative studies have shown that the amount of histamine required to produce the rate of secretion which occurs in response to a meal would be sufficient to produce a facial flush and headache. (Hanson, Grossman and Ivy, 1948). This is further evidence against a rise in the free plasma histamine being the method of stimulating acid secretion after a meal and points against gastrin being histamine.

(2) GASTRIN HAS BEEN CONSIDERED A GENERAL HISTAMINE-LIBERATOR.

This envisages the histamine free gastrin causing liberation of histamine from muscles and skin, the consequent rise in plasma histamine stimulating the parietal cells. There is little to support this concept. However, Smith (1954) injecting an almost histamine free preparation of gastrin, prepared from pigs, into the aorta of cats under chloralose anaesthesia did stimulate acid secretion and perfusion experiments demonstrated the release of histamine from skeletal muscles and skin. This histamine release could have been a reaction to the injection of the protein material contained in the injection. Indeed it seems that any compound containing two or more basic groups carried on and separated /

/separated by a sufficient aliphatic or aromatic scaffold is liable to have this property. The facts previously mentioned under the first concept, pointing against a rise in the histamine level in the general circulation being the cause of post-prandial secretion, also fail to support this conception of histamine's role.

(3) HISTAMINE IS LOCALLY RELEASED AT THE PARIETAL CELL.

The presence of histamine in the gastric juice, its high concentration in the fundus of the stomach and the absence of a demonstrable increase in the blood during digestion, led Babkin (1938) and McIntosh (1938) to suggest that it might be locally released at the parietal cell as the final step in stimulating acid secretion. This role for histamine has been supported by the observations of several workers, Emmelin and Kahlson, 1944: Code, Hallenbeck and Gregory, 1947: Grossman and Robertson, 1948. The recent demonstrations by Code (1955) of a definite correlation between secretory activity of the parietal cell and the histamine activity of the gastric juice would add further support to this concept if it could be conclusively proven that there is no rise in the free histamine in the blood during digestion.

Then there are the experiments with antihistaminic drugs. Such substances have molecules bearing some /

/some resemblance to that of histamine and presumably act by competing with histamine for receptors in the tissues. (Gaddum's Pharmacology, 1953). Antihistaminics antagonise many of the effects which are produced by histamine release so their effect on acid secretion is of interest. However, since antihistaminics produce other effects such as local irritation and anaesthesia and atropine-like actions especially in high concentration, which are not related to their antagonism to histamine, such evidence must be interpreted with care.

Most observers found antihistaminic drugs, given systemically did not inhibit acid secretion in dogs (Loew and Chichering, 1941: Burchell and Varco, 1942: Hallenbeck, 1943: Sangster, Grossman and Ivy, 1946) in man, Morrach, Rivers and Morlock, 1946, and in cats, Bouet and Walthert, 1944: Wood,1948.

However, when such histamine antagonists were applied locally to the gastric mucosa of dogs in massive concentration inhibition of acid secretion occurred. Grossman and Robertson, 1942: Kay and Forrest, 1956). Grossman and Robertson did not think, however, that there was evidence of a specific histamine-antihistamine reaction in their experiments since analogues with very weak antihistaminic action inhibited acid secretion equally. However, Kay and Forrest thought the action was specific since acid secretion alone was inhibited, pepsin secretion remaining. Thus the promethazine which they /

/they introduced into the pouches of their dogs might have blocked the action of histamine on the parietal cell without producing any effect on the pepsin secreting cells. This may well be the explanation but other possibilities arise. Promethazine in high concentration contains atropine-like effects to a greater degree than other antihistaminics and one possibility is that when applied to the mucosa at 6 mg/ml. it acts in this way. In this respect it is interesting to note that atropine does not completely abolish pepsin secretion (Linde, S., 1950). Another interesting speculation arises out of some observations on the action of histamine on pepsin secretion. As long ago as 1931 Webster found that after a subcutaneous dose of 0.5 mg. histamine, the decrease of pepsin concentration was not so pronounced and did not reach such a low level as after the administration of a larger dose (1 mg.). In pouch dogs and dogs with gastric fistulae, Alley (1935) found that the total volume of secretion in response to a test meal was reduced if a preliminary secretion had been invoked by a subcutaneous injection of histamine. The peptic response was much lowered, if histamine had been given previously. One possible explanation of those results of Webster and of Alley is that when the rate of secretion is slow some of the peptic glands are quiescent and "the washing out" effect of the pepsin weaker. However Babkin (1950), believes these results could be explained as an inhibitory action of histamine on the /

/the secretion of pepsin. This evidence led Linde to re-investigate the effect of histamine on pepsin secretion and from his evidence he concluded that histamine has an inhibitory effect on the peptic cells. The failure to suppress pepsin secretion by locally applied antihistaminic drugs may be effected by blocking this histamine inhibition of pepsin secretion. It must also be considered possible that the locally applied drug may enter the parietal and pepsin secreting cells in different concentrations. Even if the concentration were the same in both cells, it still remains a possibility that the more highly specialised parietal cell's enzyme chemistry might be more easily deranged, without any specific reaction occurring. Gregory (1955) reviewing the evidence that histamine is concerned as a local agent in the excitation of the parietal cell pointed out that the presence of histamine in the region of the parietal cells in the absence of histaminase, together with its occurrence in gastric juice whatever the stimulus suggest it may be concerned in the excitation of the parietal cell but does not constitute a proof of this. In his own words "it must be admitted that the liberation of histamine in the parietal cell might well be an effect rather than the cause of its response."

We have now reviewed briefly the existing facts which suggest that histamine may be concerned in the physiological stimulus of acid secretion and the various theories which have been developed to explain these observations. /

/observations.

The statement of Hanson, Grossman and Ivy (1948) still appears a fair summary of this evidence. "In the final analysis, no direct proof exists for the participation of histamine in the normal mechanisms for gastric secretion, either as a humoral agent or as a local chemical mediator. On the other hand, neither of these possibilities is disproven by the available evidence."

Clarification of the precise role of histamine in acid secretion has been retarded by the absence of a reliable method of estimating physiological quantities of free histamine in the plasma. Indeed it is possible that a rise in the free histamine of arterial blood, sufficient to produce acid secretion, may not be associated with a corresponding rise in the venous blood sampled, since it may be removed by one single passage through the tissues. (Code, 1955).

Although no sensitive blood method exists a new method of measuring the output of free histamine in the urine has been developed by Roberts and Adam (1951). Using this technique, these authors were able to detect a rise in the free histamine in the urine when histamine was infused intravenously at dose levels which barely stimulated the parietal cells and produced no other symptoms. When a dog had intravenous infusions of histamine at dose levels which produced an acid response equivalent to the post-prandial /

/post-prandial secretion, the output of free histamine in the urine was greatly elevated, levels often more than ten times that of the control period being recorded (Updike, 1955). Here then was a method of indirectly measuring the level of free histamine in the plasma, quite sensitive enough to record changes previously only detectable by the parietal cell response. It suggested that an examination of the possible relationship betw een the appearance of free histamine in the urine and secretion of acid by the gastric mucosa might prove profitable in elucidating the role of histamine in acid secretion.

In the following chapter previous knowledge concerning the excretion of histamine in the urine will be briefly outlined.

The information collected by other authors using the recent method of Roberts and Adam will then be reviewed.

Data collected using this method to study the changes in the output of free histamine in the urine during digestion will then be presented in the chapters which follow.

CHAPTER II.

URINARY HISTAMINE

INTRODUCTION.

It is only in recent years that the presence of histamine in the urine has been demonstrated. Earlier workers using biological methods (Dale and Laidlaw, 1910: Oehma, 1913: Guggenheim and Loeffler, 1916) did not detect it in the urine of animals even after injection of large amounts of histamine.

However, in 1944, G.V.Anrep and others reported their results after studying the urine of different animals in the Cairo Zoo. They found that extracts of normal urine prepared by a modified technique of Barsoum and Gaddum (1935) were inactive when tested for histamine unless they had previously been hydrolysed in acid. After hydrolysis in hydrochloric acid the extracts showed unmistakeable evidence of containing histamine. As well as this inactive "conjugated histamine" which on hydrolysis produced active histamine, small amounts of free histamine were also detected in the urine on some occasions. They found that herbivora excreted histamine mainly in the free form, carnivora mainly in the conjugated form. Rats occupied an intermediate position, as some 40 - 50% of the histamine excreted in the urine occurred in /

/in the free form.

The effect of diet on the excretion of histamine in the urine was clearly demonstrated by these workers. Thus rats, while on a carbohydrate fat diet, had very low levels of urinary histamine (below 10 mg/day) and most of it was in the free form. When maintained on a meat diet the total histamine in the urine rose to over 90 mg/day. Although the free histamine in the urine nearly doubled on a meat diet, most of the increase was accounted for by a rise in the output of conjugated histamine. Furthermore this increased output of histamine in the urine did not occur on a high protein diet devoid of meat (casein, cheese and egg albumen). Rather similar findings were obtained in dogs and man.

Anrep and his co-workers also studied the effect of administering histamine itself. When histamine was injected subcutaneously in dogs a rapid rise in the levels of free histamine occurred. This was shown to be also true in man (R. Mitchell, 1956). On the other hand, when histamine was administered to dogs by mouth, the conjugated form increased rapidly in the urine, the peak of excretion occurring 7-14 hours later. Anrep's group thus clearly defined some of the factors controlling the appearance of histamine in the urine and drew attention to the fact that it occurred in both free and conjugated forms. /

/forms.

CONJUGATED HISTAMINE IN THE URINE.

This study is not concerned with conjugated histamine. It will be sufficient to note that its chemistry and site of formation have been established. Rosenthal and Taylor noticed certain similarities between conjugated histamine and acetyl-histamine. Urbach demonstrated the identity of the migration rates of the inactive form of histamine in the urine and synthetic acetyl-histamine. Finally Tabor and Mossetig isolated from the urine of dogs fed histamine, acetyl-histamine which was crystallised, characterised and identified. In addition, Urbach suggested that histamine in the alimentary tract was converted to acetyl-histamine by the intestinal bacteria and that some of the conjugated histamine was absorbed and excreted in the urine unchanged. This theory was supported by the fact that acetyl-histamine was formed from histamine added to normal, but not to autoclaved, stools. The possibility that the liver played a part in the acetylation of histamine could not be excluded and this was investigated by Livingstone and Code. Histamine was given into the portal veins of experimental animals. The urinary excretion of free and conjugated histamine was measured before, after and during infusion. Evidence that the liver conjugated histamine was not obtained in dogs or monkeys. Acetyl-histamine may not be the only form of inactive histamine occurring in /

/in the body. This will be discussed in a later chapter.

FREE HISTAMINE IN THE URINE.

Further studies on the excretion of free histamine in the urine were for some time retarded by the absence of a sensitive method of measuring it. However, in 1950, Roberts and Adam reported their technique of estimating free histamine in body fluids. After investigating a number of commercially available cation exchangers, a synthetic zeolite Decalso was used for the absorption of free histamine from body fluids and its subsequent elution was in a state pure enough for pharmacological assay. This method has been applied to the study of free histamine in the urine by several authors.

It was found that when histamine was given intravenously, the amount of free histamine excreted in the urine increases. If, at the same time, gastric juice was collected, a correlation was found between the output of acid by the gastric mucosa and the output of free histamine in the urine, both in turn being related to the amount of histamine injected (Adam, Card, Riddell, Roberts and Strong, 1954).

The effect of diet on the excretion of free histamine was re-examined with this method. When a diet high in meat was fed human beings the excretion of free histamine in the urine increased decisively above fasting levels and /

/and levels obtained when diets free of meat were eaten (Mitchell and Code, 1954).

Now a meat meal is one of the most potent stimuli to the secretion of acid in the gastric juice, and Updike, working in Code's laboratory at Rochester, found a rough parallelism between the hourly output of free histamine in the urine and the hourly output of hydrochloric acid in the juice secreted from Heidenhain gastric pouches of dogs fed a meal of meat.

The present study was undertaken to examine further the possible relationship between the appearance of free histamine in the urine and secretion of acid by the gastric mucosa, particularly during stimulation following the ingestion of a meat meal.

CHAPTER III.

EXPERIMENTAL PLAN, MATERIAL AND METHODS.

EXPERIMENTAL PLAN.

The possible relationship between the appearance of free histamine in the urine and acid secretion by the gastric mucosa was first studied.

The parallelism noted by Updike and others (1955) between output of acid by dogs with Heidenhain pouches and excretion of free histamine in the urine after a meal of meat was first confirmed (Chapter IV).

Since the free histamine appeared in the urine only during acid secretion, the possibility that it derived from the parietal cell region could not be excluded. Therefore similar studies were carried out in totally gastrectomised dogs (Chapter V).

An effort was then made to separate the acid secretory response into vagal, gastric and intestinal components and to determine if there was any relationship between free urinary histamine and free acid in the gastric juice during any of these phases (Chapters VI, VII, and VIII).

The above studies appeared to demonstrate that the free histamine occurring in the urine after a meal of meat was not related to acid secretion. It only occurred when /

/when the meat entered the small bowel and was peculiar to meat. The possibility that the presence of L-histidine in the meat might prove a substantial source of the urinary histamine was studied further (Chapter IX).

The above studies suggested that intestinal bacteria, by decarboxylating L-histidine to histamine, might play an important part in the augmentation of urinary histamine by a meat meal. Would histamine, formed in this way in the lumen of the alimentary tract, be absorbed? To answer this question, studies of histamine absorption from the small bowel were carried out (Chapter X).

MATERIAL AND METHODS.

(a) DOG STUDIES.

Mongrel dogs of 9-15 Kg. body weight were used. Nearly all were females to facilitate catheterisation of the urethra. The animals were in good health throughout the study, their weight and appetite being maintained.

OPERATIVE PROCEDURES.

These were performed with the animals under ether anaesthesia using a strictly aseptic technique. The animal preparation used in each study will be described in the appropriate chapter.

(b) HUMAN STUDIES.

These were carried out in adult patients /

/patients convalescing from appendicectomy or herniorrhaphy.

(c) CONDITIONS OF TESTS.

a) Dogs. All animals were fasted for 36 hours before each test. Tests consisted of simultaneous hourly collections of urine and gastric juice before and after administration of a meal or insulin. The content of the meal and its mode of administration will be described in the individual studies.

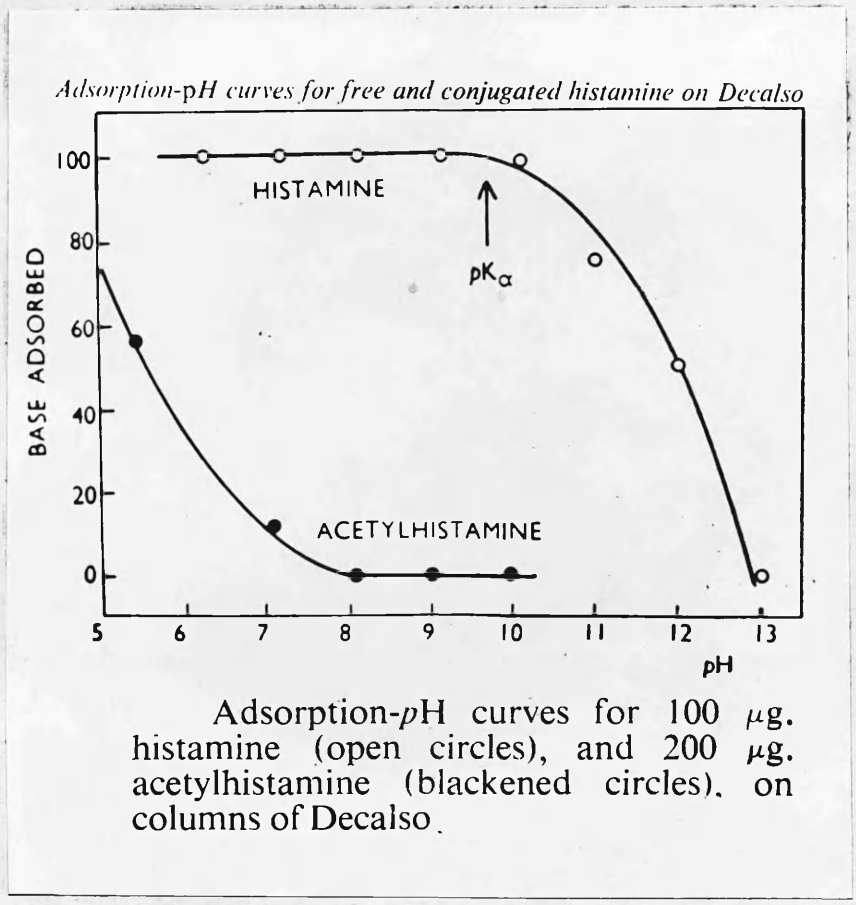
To provide adequate volumes of urine for histamine determinations all animals had continuous I.V. infusions of 0.45% saline solution throughout the entire period of the tests. Urine was collected continuously from indwelling Foley catheters. Each hourly urine sample was acidified and refrigerated to preserve its free histamine content.

b) Human Studies. Tests on patients followed a 16 hour fast. Urine was collected by hourly voiding and an adequate urinary flow obtained by oral fluids.

ESTIMATION OF FREE HISTAMINE IN THE URINE.

The Decalso method described by Roberts and Adam was used to separate the free histamine from the urine. Such adsorbents act by cation-exchange and are capable of taking up the ions of organic bases from very dilute solutions. The synthetic zeolite known as Decalso adsorbs only free histamine in the pH range 8 - 10. (Fig. I) Glass columns /

/columns containing Decalso were prepared as illustrated in Fig. II.



Adsorption-*p*H curves for 100 μg. histamine (open circles), and 200 μg. acetylhistamine (blackened circles), on columns of Decalso.

FIG. I.

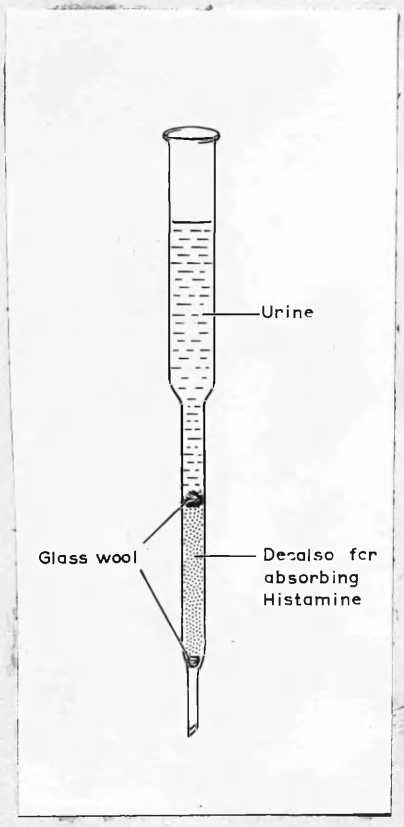


FIG. II.

Each column contained about 3 gms. of the zeolite. It was carefully packed into place with a glass plunger to achieve a density allowing 50 ml. of urine to percolate through the column in not less than 1 hour. Each hourly collection of urine was adjusted to a pH of 8using Universal indicator, filtered and 50 ml. placed on a Decalso column which had previously been moistened by 10 ml. distilled water. Where the hourly output of urine was less than 50 ml. it was made /

/made up to this volume before being placed on the column. Following the urine, each column was washed with 25 ml. of normal saline and most of the water removed by 15 ml. absolute alcohol. The free histamine was eluted with 3.5 ml. AnalaR ammonium hydroxide, followed by 50 ml. of ammoniated chloroform, this organic solvent carrying the liberated histamine through the column and into a 300 ml. pressure flask. The chloroform ammonia eluate in each flask was then evaporated to dryness in a water bath at 40°C under reduced pressure. The slight residue was then dried off with 10 ml. absolute alcohol containing 3% (V/V) concentrated HCl to neutralise traces of alkali and finally with 10 ml. of absolute alcohol. The acidified extract could be stored in a dessicator for several days without loss of activity.

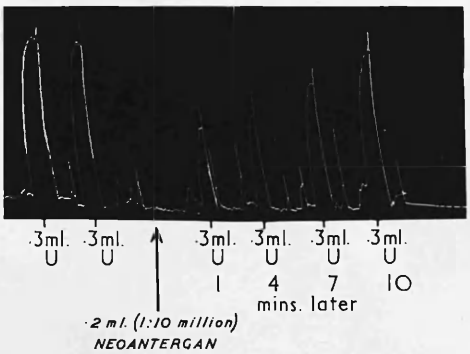
Finally extracts were taken up in known volumes of saline, their pH adjusted to 7.5 using a pH meter and the histamine content estimated by bio-assay, using strips of guinea-pig ileum, against known standard solutions of histamine. From this, the hourly output of free histamine could be calculated. In each experiment, in addition to a Decalso column for each hourly urine collection, known amounts of histamine were added to 50 ml. of urine or saline and passed through prepared columns to measure the efficiency of the extraction method. The original authors, Roberts and Adam found 67$\pm$ 1% of free histamine added to urine was recovered. /

Fig. 2a.

EFFECT OF NEOANTERGAN ON CONTRACTIONS OF GUINEA PIG ILEUM INDUCED BY—

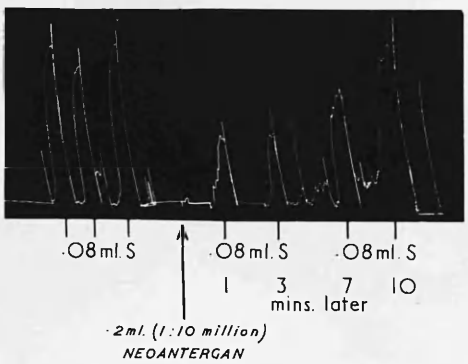
1. SUBSTANCE IN URINE CONSIDERED TO BE HISTAMINE.
2. STANDARD SOLUTION OF HISTAMINE.

1.



U—Substance in urine considered to be Histamine.

2.



S — Standard solution of Histamine.

/recovered.

In 102 control tests, in which histamine was added to 50 ml. of urine or saline in the range 0.5 to 3 ug. a lower mean recovery of free histamine was found and the extraction method in my hands showed a greater variation in recovery rate (Mean Recovery + 59.9% $\pm$ 3.4 S.E. of mean).

The urinary histamine results are all expressed as the output of histamine base in the urine per hour and no correction has been made for the fraction of histamine lost in the extraction process.

TESTS TO IDENTIFY THE URINARY FREE HISTAMINE.

(1) ANTIHISTAMINICS. Small doses of the antihistaminic drug, promethazine, when added to the organ bath inhibit the response to a dose of standard histamine solution by a definite amount and a definite time elapses before the gut again gives a full contraction to that dose of histamine. The same amount of this antihistaminic inhibits an equivalent contraction of the gut produced by the urinary extract by the same degree and an identical time is required for complete recovery of the gut's response. (Fig. IIa - on opposite age.)

This distinguishes histamine from many other substances, but not from N-methyl histamine (Schild, 1947). The latter compound would, however, give discordant results when tested against histamine by parallel assays on guinea pig gut and cat blood pressure (Vartiginen, 1935). Present methods of parallel assay would probably not differentiate histamine from N-dimethyl histamine. (Gaddum, 1948).

(a) HISTAMINASE DESTROYS THE URINARY SUBSTANCE. This is further confirmatory evidence that the substance in the urine is histamine. However, little is known about the action of this enzyme on pharmacologically active compounds closely related to histamine such as the N-alkyl and N-dialkyl histamines (Roberts and Adam, 1950).

FREE ACID IN THE GASTRIC JUICE.

The hydrochloric acid in the gastric juice was determined by titration with N/10 NaOH using dimethyl amino-benzene as indicator and from this titration the hydrochloric acid output per hour calculated and expressed in milliequivalents.

CONTROL EXPERIMENTS.

The different studies reported in the following chapters frequently required special control experiments which differed from the general experimental plan outlined here. Details of these separate control studies will be described in the appropriate chapters.

CHAPTER IV.

ACID SECRETION AND OUTPUT OF FREE HISTAMINE IN THE URINE FOLLOWING A MEAT MEAL BY MOUTH.

It will be remembered that previous workers had found that when a diet high in meat is fed rats or human beings, the excretion of free histamine in the urine increases decisively above fasting levels and levels obtained when diets free of meat are eaten. What is the significance of the free histamine occurring in the urine after the meat has been eaten? Does it indicate that histamine is concerned in some way in the acid response to the meal? Certainly the rough parallelism of acid response and urinary histamine noted by Updike et al (1955) suggested that the two might be related. In the present chapter an endeavour was made to confirm their observations.

OBJECT OF EXPERIMENTS .

To relate the hourly output of free histamine in the urine following a meal of meat administered orally to dogs, to the acid response obtained from denervated gastric pouches of Heidenhain type.

MATERIAL AND METHODS.

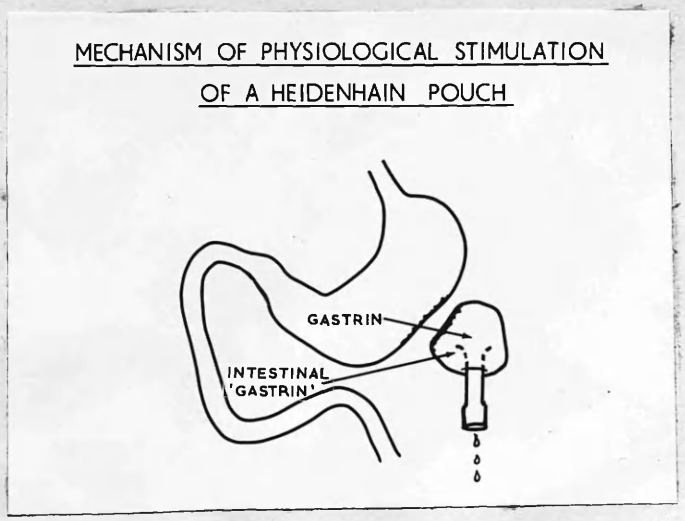
These experiments were performed on healthy female /

/female mongrel dogs of 9-15 kilo body weight, all of which had separated denervated gastric pouches of Heidenhain type (Fig. 3).

The meat meal consisted of 170 gms. of cooked horse meat, the source of meat being the same in all experiments. Each experiment was performed after the animal had been fasted 36 hours. A self-retaining 5 ml. Foley catheter was inserted into the bladder with the use of a vaginal speculum, under aseptic conditions, and the bladder washed out. The dog was then placed on a suitable stand and an intravenous drip of hypotonic saline commenced to ensure a continuous output of urine. Hourly collections of gastric juice from the pouch and urine from the bladder were then started. After a control period of 1 or 2 hours, a meat meal consisting of 170 gms. of cooked horse meat was given by mouth and the hourly collections continued for a further 8 - 10 hours.

The hourly output of acid by the gastric pouch and histamine in the urine was then obtained by the methods previously described.

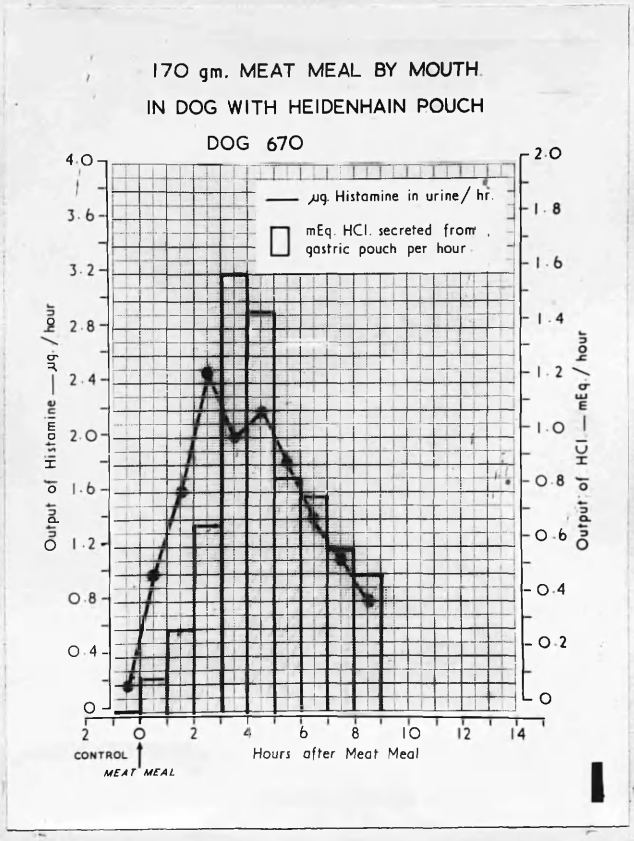
Fig. 3.



RESULTS.

Five tests were carried out on four dogs. The results are illustrated below:-

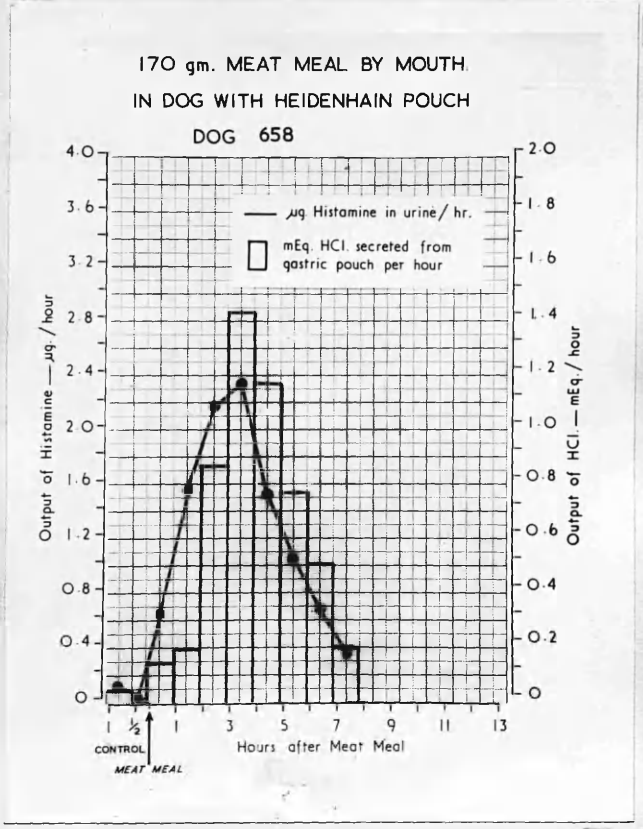
Fig. 4



EXP	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE μg.	VOL. GASTRIC JUICE SECRETED ml / Hr.	mEq. HCl per hr.
670	CONTROL HOUR	48	ALL	10	0·4	0·08	0·2	0	0
	HOUR 1	36	"	"	0·2	0·2	1·0	1·0	0·12
	HOUR 2	50	"	"	0·12	0·2	1·6	3·0	0·3
	HOUR 3	38	"	"	0·08	0·2	2·5	5·6	0·37
	HOUR 4	40	"	"	0·1	0·2	2·0	11·0	1·6
	HOUR 5	28	"	"	0·09	0·2	2·2	12·2	1·45
	HOUR 6	35	"	"	0·11	0·2	1·8	7·1	0·85
	HOUR 7	40	"	"	0·14	0·2	1·4	7·0	0·77
	HOUR 8	46	"	"	0·18	0·2	1·1	6·2	0·59
	HOUR 9	50	"	"	0·23	0·2	0·8	5·5	0·49
	HOUR 10								

HISTAMINE STANDARD — 0·1 μg. / ml.

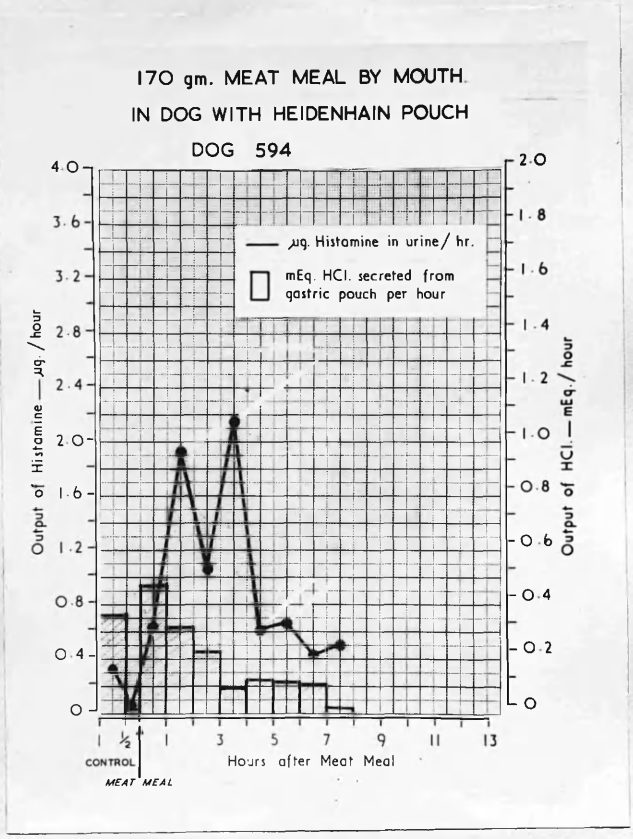
Fig. 5



EXP	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE μg.	VOL GASTRIC JUICE SECRETED ml / hr	mEq HCl per hr
658	CONTROL HOUR 1	40	40	10	0·3	0·04	0·13	1·2	0·04
	CONTROL 1/2 HR	13	13	"	0·4	0·02	0·05	0·2	0·01
	HOUR 1	108	50	"	0·16	0·04	0·548	2·2	·143
	HOUR 2	66	50	"	0·04	0·04	1·554	2·3	·195
	HOUR 3	109	50	"	0·04	0·04	2·18	7·5	·85
	HOUR 4	117	50	"	0·04	0·04	2·34	11·2	1·4
	HOUR 5	75	50	"	0·04	0·04	1·32	9·2	1·18
	HOUR 6	70	50	"	0·08	0·06	1·05	5·0	0·77
	HOUR 7	95	50	"	0·14	0·05	0·684	4·5	0·51
	HOUR 8	71	50	"	0·15	0·04	0·37	2·0	0·2
	HOUR 9								

HISTAMINE STANDARD — 0·1 μg. / ml.

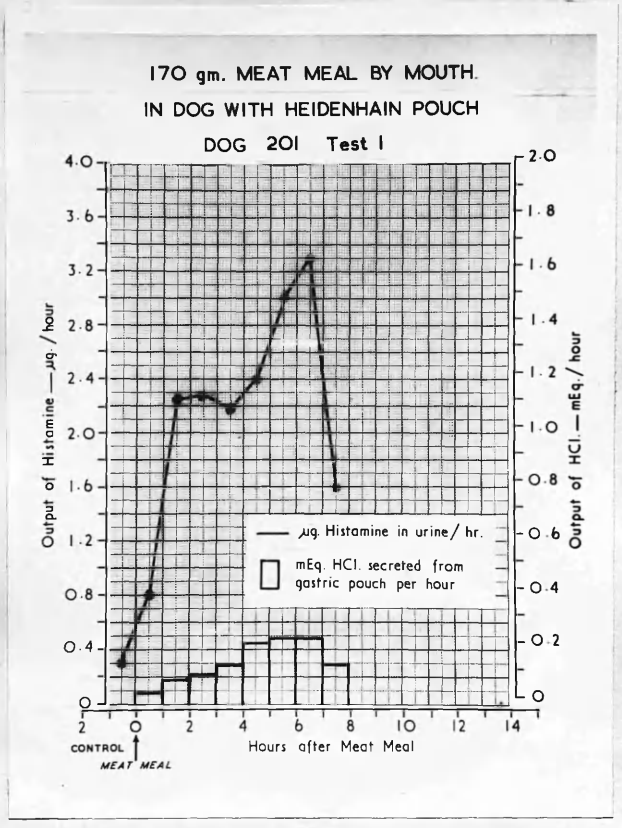
Fig. 6



EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE μg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl per hr.
594	CONTROL HOUR 1	40	40	10	0·3	0·1	0·33	4·8	·36
	CONTROL ½ HR	59	59	"	0·5	0·03	0·06	0·3	·019
	HOUR 1	200	50	"	0·3	0·05	0·64	4·5	·47
	HOUR 2	185	50	"	0·19	0·1	1·95	3·1	·32
	HOUR 3	210	50	"	0·4	0·1	1·05	2·3	·22
	HOUR 4	150	50	"	0·28	0·2	2·16	1·0	·09
	HOUR 5	160	50	"	0·5	0·09	0·6	1·7	·12
	HOUR 6	165	50	"	0·5	0·1	0·66	1·6	·12
	HOUR 7	120	50	"	0·5	0·09	0·43	1·4	·11
	HOUR 8	50	50	"	0·2	0·1	0·5	0·6	·02
	HOUR 9								

HISTAMINE STANDARD — 0·1 μg. / ml.

Fig. 7



EXP	SPECIMEN	TOTAL URINE VOL. ml	URINE VOL. ON DECALSO COLUMN ml	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN ml	HOURLY URINARY HISTAMINE µg	VOL GASTRIC JUICE SECRETED ml / hr	mEq HCl per hr
2 C 1 TEST I	CONTROL HOUR	40	ALL	10	0·4	0·2	0·3	10	NONE
	HOUR 1	34	ALL	"	0·1	0·08	0·8	10	0·04
	HOUR 2	90	50	"	0·02	0·1	2·25	15	0·09
	HOUR 3	115	"	"	0·12	0·3	2·28	16	0·1
	HOUR 4	82	"	"	0·06	0·08	2·18	24	0·18
	HOUR 5	102	"	"	0·1	0·12	2·4	33	0·22
	HOUR 6	100	"	"	0·08	0·12	3·0	38	0·24
	HOUR 7	125	"	"	0·1	0·1	3·2	38	0·24
	HOUR 8	50	"	"	0·02	0·1	1·6	26	0·14
	HOUR 9								
	HOUR 10								

HISTAMINE STANDARD — 0·1 µg / ml.

Fig. 8

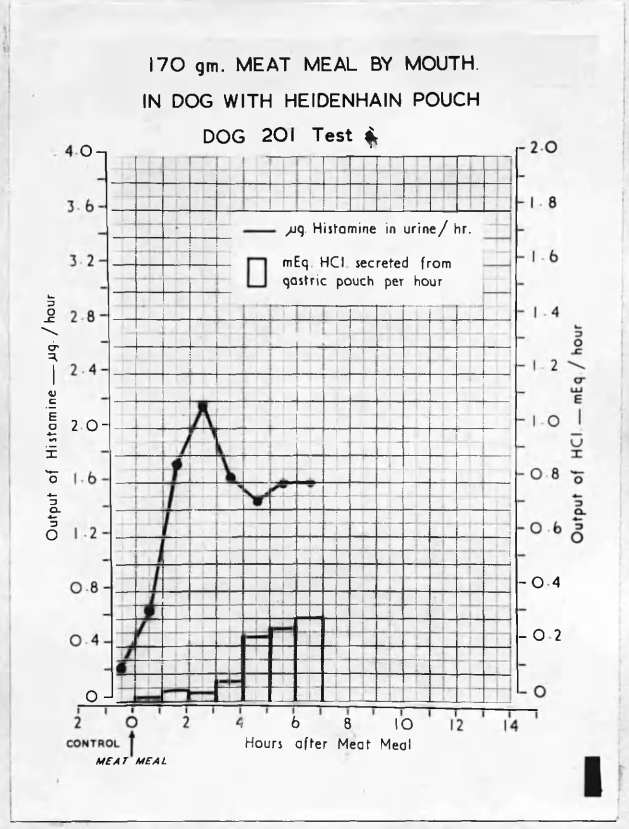


Fig. 9

EXP	SPECIMEN	TOTAL URINE VOL. ml	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL. UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE μg.	VOL. GASTRIC JUICE SECRETED ml / hr.	mEq HCl per hr.
201 Test 2	CONTROL HOUR 1	50	50	10	0·4	0·1	0·25	1·6	0
	CONTROL HOUR 2	50	"	"	0·12	0·08	0·66	1·5	0·02
	HOUR 1	1·0	"	"	0·1	0·08	1·74	1·2	0·04
	HOUR 2	82	"	"	0·06	0·08	2·18	1·2	0·03
	HOUR 3	62	"	"	0·06	0·08	1·65	2·0	0·07
	HOUR 4	55	"	"	0·06	0·08	1·45	2·5	0·23
	HOUR 5	50	"	"	0·05	0·08	1·5	3·1	0·25
	HOUR 6	50	"	"	0·05	0·08	1·6	4·5	0·3
	HOUR 7								
	HOUR 8								
	HOUR 9								

HISTAMINE STANDARD — 0·1 μg. / ml.

COMMENTARY ON RESULTS.

The results obtained in these experiments reveal a very similar pattern. During the control period of 1 - 2 hours, before the meal was given, the output of free histamine in the urine was always low and relatively fixed at levels ranging between 0.15 - 0.35 ug per hour. The output rose rapidly following ingestion of the meat and in the second, third and fourth hours had usually reached 2 ug/hour or more. The level of histamine in the urine then fell very gradually and low levels, close to those obtained in the control period, were only obtained nine or ten hours after feeding. This is an identical pattern to that found by Updike and co-workers. As in their experiments, there was a rough parallelism between the output of acid from the pouch and the output of free histamine in the urine. Both reached maximal values between the 2nd and 6th hour after the meal and then gradually declined towards control levels (Figs. 4 - 9).

This simultaneous rise and fall of histamine and acid secretion is well illustrated when the mean results of the 5 tests on 3 dogs is expressed graphically (Fig. 10.)

/

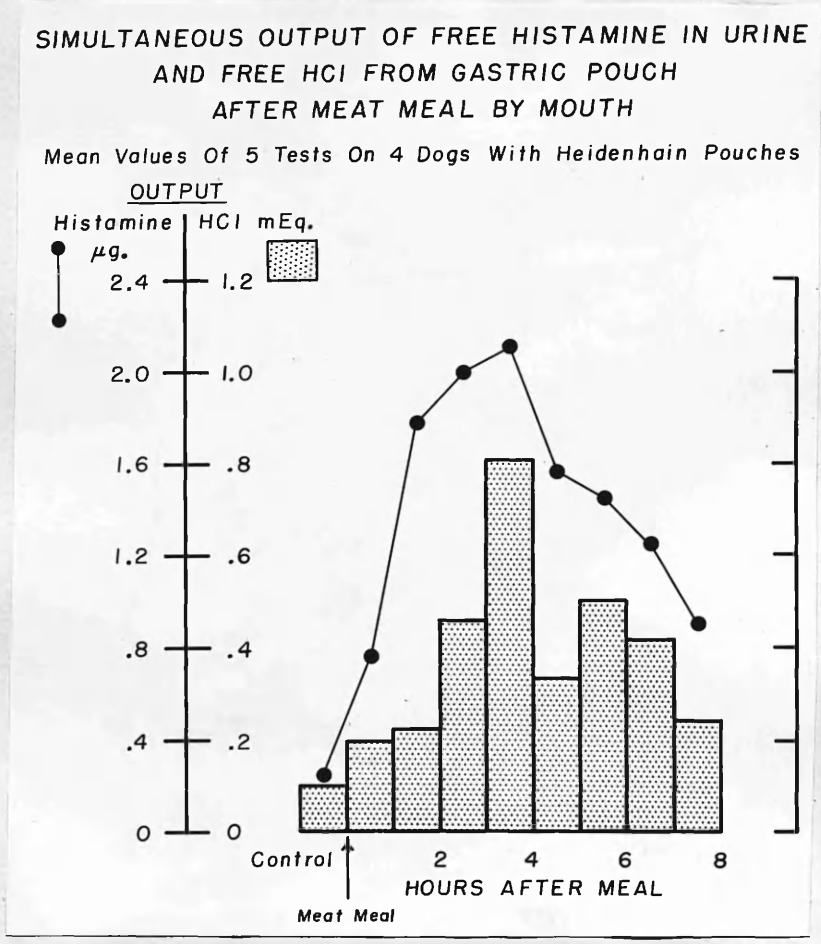


FIG. 10

Although this parallelism between output of acid and histamine occurred no very close correlation existed between the urinary histamine and acid secretion, hour by hour. The degree of correlation found in two of the dogs is illustrated graphically in Figs. 11 and 12. When the /

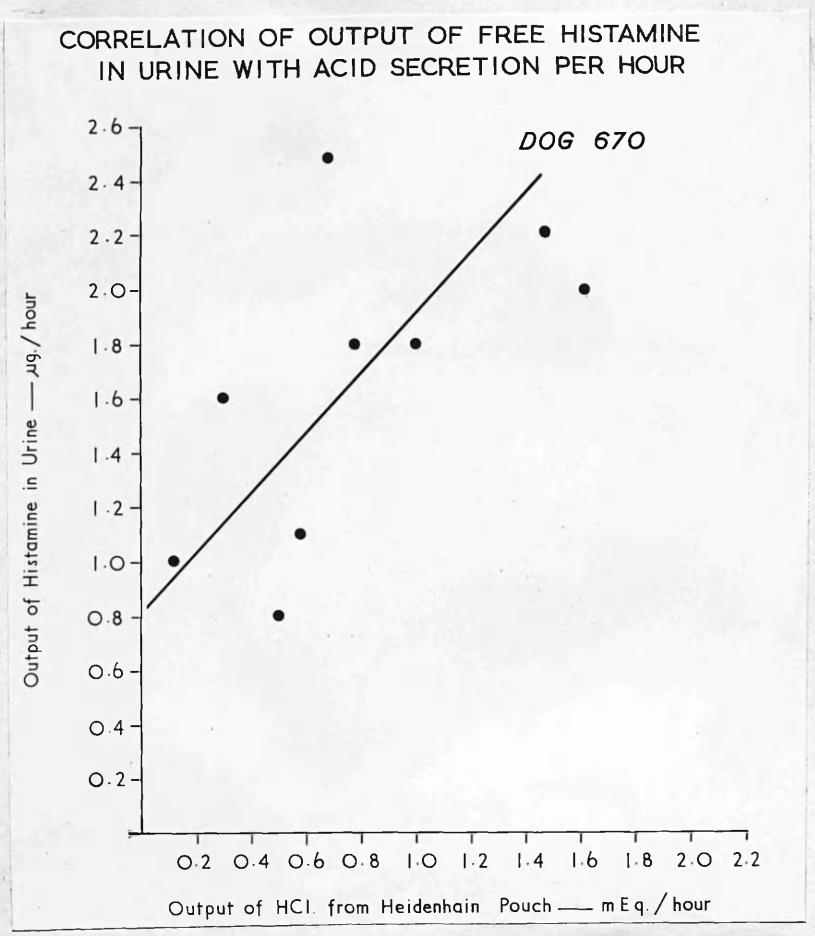

CORRELATION OF OUTPUT OF FREE HISTAMINE
IN URINE WITH ACID SECRETION PER HOUR
DOG 670
Output of Histamine in Urine — µg./hour
2·6
2·4
2·2
2·0
1·8
1·6
1·4
1·2
1·0
0·8
0·6
0·4
0·2
0·2 0·4 0·6 0·8 1·0 1·2 1·4 1·6 1·8 2·0 2·2
Output of HCl from Heidenhain Pouch — m Eq./hour

Fig. 11

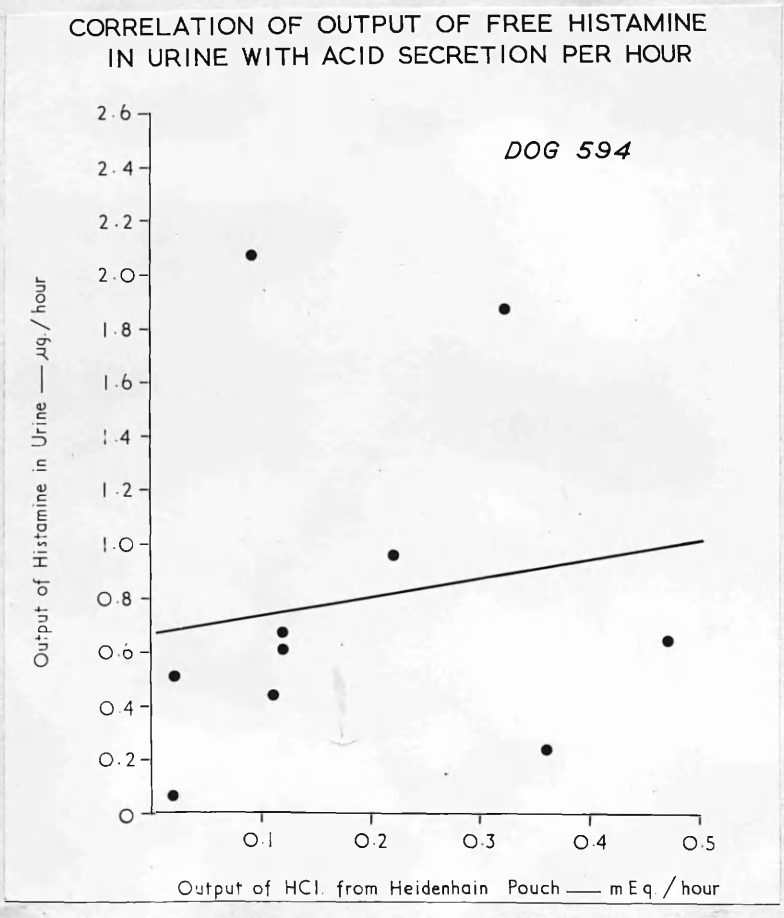

CORRELATION OF OUTPUT OF FREE HISTAMINE
IN URINE WITH ACID SECRETION PER HOUR
DOG 594
Output of Histamine in Urine — µg./hour
2·6
2·4
2·2
2·0
1·8
1·6
1·4
1·2
1·0
0·8
0·6
0·4
0·2
0
0·1 0·2 0·3 0·4 0·5
Output of HCl from Heidenhain Pouch — m Eq./hour

Fig. 12

/the relationship of the urinary histamine to acid secretion is studied statistically taking in all 46 hourly collections in the 5 tests, a significant association between the two was found. The analysis of covariance is given below.

ANALYSIS OF COVARIANCE.

Source of Variation	Sum of Squares	Degrees of freedom	Mean Square
Overall regression coefficient	V_o=9.3	1	9.34**
Differences among regression coefficients for individual dogs	V_1=2.45	4	0.61 (N.S)
Deviations of dog means from their regression line	V_2=5.162	3	1.72*
Regression coefficient for dog means	V_3=0.190	1	0.19 (N.S)
Residual	V_4=13.74	36	0.38
TOTAL	30.88	45	

CONCLUSIONS OF COVARIANCE

(i) Histamine and gastric juice are associated.

(ii) There are no differences in slopes of regression lines of histamine and gastric juice for different dogs.

(iii) Overall correlation coefficient = 0.6

These results posed two separate questions. Firstly the acid response in these studies involved both the gastric and intestinal phases of acid secretion. Would separation of these two humoral components reveal a closer correlation between acid output and urinary histamine? Secondly, what was the probable source of the histamine in the urine? Was it derived from the parietal cells having been absorbed into the general circulation after being locally released by these humoral mechanisms? Again, was the histamine release in the urine related to absorption of histamine itself or a general histamine liberator? In short, was gastrin histamine or a general histamine liberator?

There remained another quite different approach to interpretation of these results. Was the rise in urinary histamine, after meat is ingested, unassociated with a rise in blood histamine? If this were so, then the formation of a histamine releasor having its effect in the kidney alone must be considered.

In the studies which follow, an attempt has been made to investigate some of these possibilities.

CONCLUSIONS.

(1) After a meat meal by mouth, there is a parallelism between the output of histamine in the urine and acid secretion from denervated pouches of Heidenhain type.

(2) In individual dogs there is a moderate but /

/butsignificant correlation between acid secretion and free histamine output in the urine, hour by hour, when the different phases of acid secretion are not separated. (Overall correlation coefficient = 0.6).

CHAPTER V.

THE EFFECT OF A MEAT MEAL BY MOUTH AFTER TOTAL GASTRECTOMY.

In the previous chapter it was noted that in dogs there was a rough parallelism between the hourly output of free histamine in the urine and the output of acid from vagally denervated gastric pouches, following a meal of meat.

Since the raised output of free histamine in the urine occurred only during gastric secretion, it was considered possible that it might be derived from the gastric mucosa. Thus, if histamine were locally released at the parietal cell during stimulation of the gastric mucosa, some might gain entrance to the general circulation during gastric secretion, with consequent overflow into the urine during this period.

OBJECT OF STUDY.

To measure the hourly output of histamine in the urine of totally gastrectomised dogs following a meal of meat by mouth.

MATERIAL AND METHODS.

A series of 15 dogs were submitted to total gastrectomy, restoration of alimentary continuity being /

/being achieved by oesophago-duodenal anastomosis. Only six of these animals permanently recovered from this procedure and of these only three regained 80 - 100% of their former weight and were considered suitable for this study.

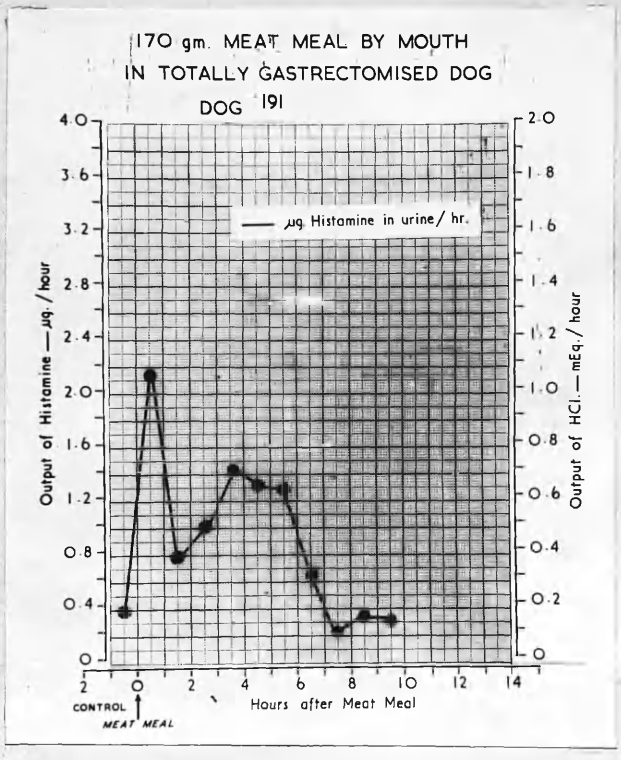
Each dog was given a meat meal of 170 gms. cooked horse meat from the same source as that used in the previous study. Urine was collected at hourly intervals before and after the meal using indwelling catheters, as previously described. No acid was collected as these dogs had no pouches. The hourly output of histamine was estimated as formerly.

RESULTS. Five tests were carried out on three dogs. The results are illustrated below:-

Fig. 13
(Dog 191)

EXP.	SPECIMEN	TOTAL URINE VOL. ml	URINE VOL. ON DECALSO COLUMN ml	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL. UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN. ml	HOURLY URINARY HISTAMINE µg	VOL GASTRIC JUICE SECRETED ml / hr	mEq HCl per hr
191	CONTROL HOUR	100	50	10	0·4	0·08	0·4		
	HOUR 1	170	45	"	0·28	0·16	2·15		
	HOUR 2	18	9	"	0·30	0·12	0·8		
	HOUR 3	110	50	"	0·24	0·12	1·1		
	HOUR 4	230	50	"	0·32	0·1	1·42		
	HOUR 5	300	50	"	0·35	0·08	1·38		
	HOUR 6	272	50	"	0·34	0·08	1·3		
	HOUR 7	100	50	"	0·26	0·08	0·62		
	HOUR 8	87	50	"	0·35	0·08	0·23		
	HOUR 9	55	50	"	0·24	0·12	0·55		
	HOUR 10	27	27	"	0·22	0·08	0·36		

HISTAMINE STANDARD — 0·1 µg. / ml.



Dog 97

Fig. 14

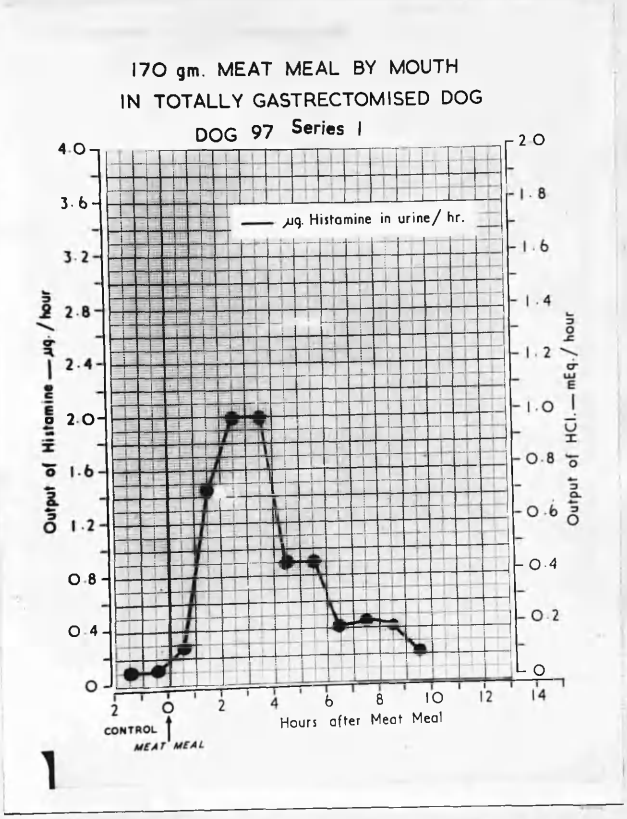
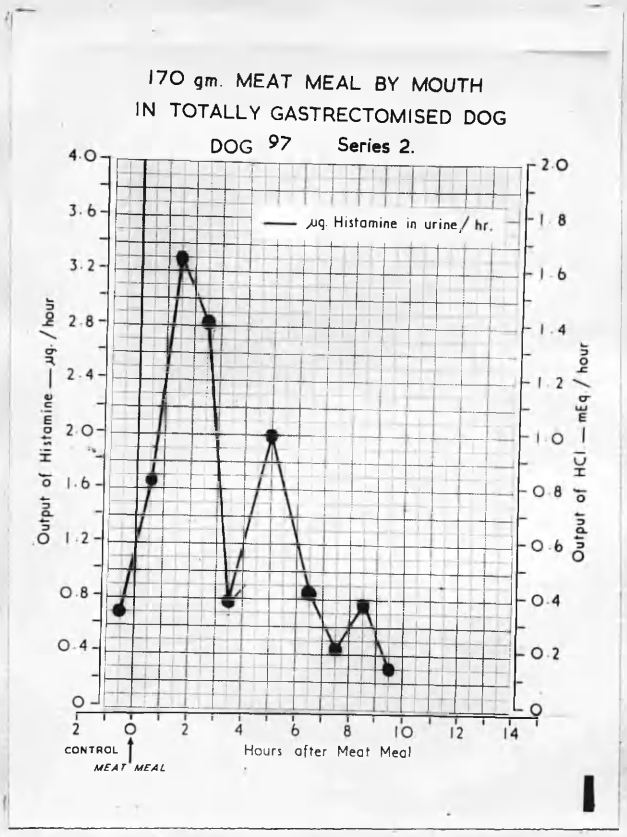


Fig. 15



EXP.	SPECIMEN	TOTAL URINE VOL ml	URINE VOL ON DECALSO COLUMN ml	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN ml	HOURLY URINARY HISTAMINE μg	VOL. GASTRIC JUICE SECRETED ml / hr	mEq HCl per hr
97 1	CONTROL HOUR 1	30	30	10	0·4	0·1	0·25		
	CONTROL HOUR 2	41	41	"	0·36	0·1	0·28		
	HOUR 1	22	22	"	0·07	0·1	1·43		
	HOUR 2	57	57	"	0·05	0·1	2·0		
	HOUR 3	48	48	"	0·05	0·1	2·0		
	HOUR 4	110	50	"	0·13	0·1	0·9		
	HOUR 5	217	50	5	0·24	0·1	0·9		
	HOUR 6	66	50	10	0·24	0·1	0·42		
	HOUR 7	18	18	"	0·22	0·1	0·46		
	HOUR 8	26	26	"	0·24	0·1	0·42		
	HOUR 9				0·4	0·08	0·2		

HISTAMINE STANDARD — 0·1 μg. / ml.

EXP.	SPECIMEN	TOTAL URINE VOL. ml	URINE VOL ON DECALSO COLUMN ml	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN ml	HOURLY URINARY HISTAMINE μg	VOL. GASTRIC JUICE SECRETED ml / hr	mEq HCl per hr
97 2	CONTROL HOUR	22	22	10	0·14	0·1	0·71		
	HOUR 1	36	36	10	0·06	0·1	1·66		
	HOUR 2	115	50	10	0·1	0·1	3·3		
	HOUR 3	185	41	10	0·16	0·1	2·82		
	HOUR 4	62	50	10	0·16	0·1	0·78		
	HOUR 5	355	44	10	0·2	0·1	2·01		
	HOUR 6	72	50	10	0·17	0·1	0·85		
	HOUR 7	36	36	10	0·23	0·1	0·44		
	HOUR 8	41	41	10	0·13	0·1	0·77		
	HOUR 9	23	23	10	0·34	0·1	0·3		
	HOUR 10								

HISTAMINE STANDARD — 0·1 μg. / ml.

Dog 105

Fig. 16

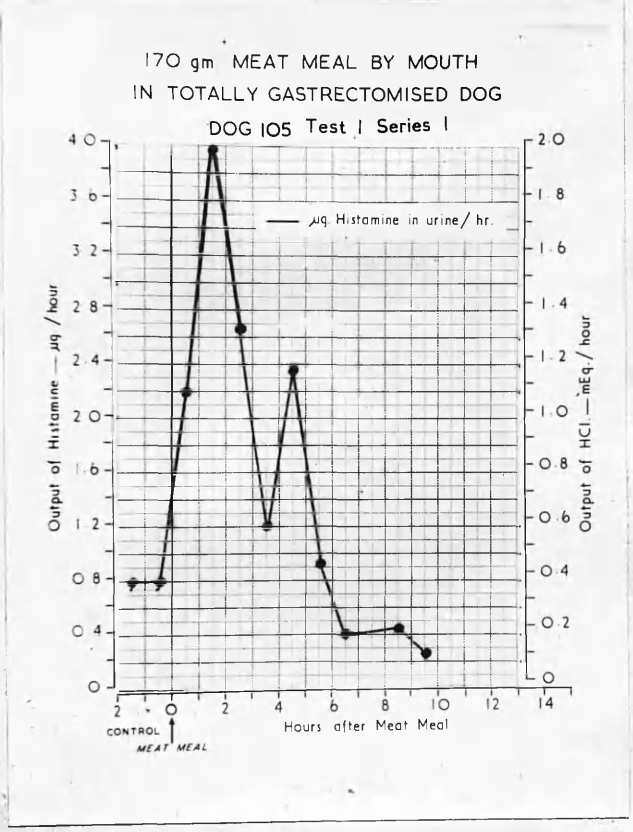
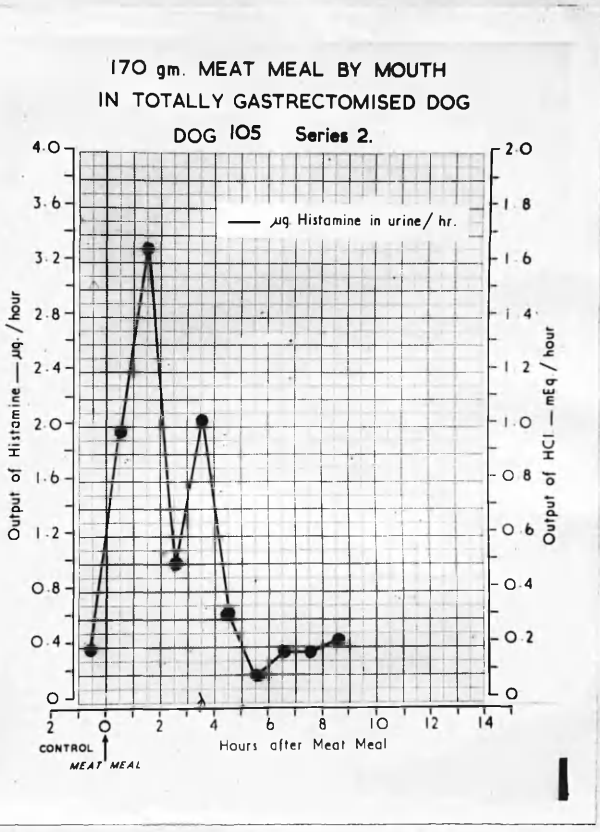


Fig. 17



EXP	SPECIMEN	TOTAL URINE VOL ml	URINE VOL ON DECALSO COLUMN ml	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL UNKNOWN ml	BIO-ASSAY HISTAMINE ST SOLN ml	HOURLY URINARY HISTAMINE μg	VOL GASTRIC JUICE SECRETED ml/hr	mEq HCl per hr
105 Test 1	CONTROL HOUR 1	30	ALL	10	0·4	0·3	0·75		
	CONTROL HOUR 2	42	ALL	20	0·05	0·06	2·2		
	HOUR 1	36	ALL	30	0·06	0·03	3·99		
	HOUR 2	125	50	10	0·04	0·085	2·65		
	HOUR 3	120	100	10	0·08	0·08	1·2		
	HOUR 4	472	100	10	0·12	0·06	2·36		
	HOUR 5	382	100	10	0·2	0·04	0·92		
	HOUR 6	150	92	10	0·2	0·04	0·40		
	HOUR 7	90	50	LOST					
	HOUR 8	40	40	10	0·2	0·09	0·45		
	HOUR 9	30	30	10	0·2	0·05	0·25		

HISTAMINE STANDARD — 0·1 μg./ml.

EXP	SPECIMEN	TOTAL URINE VOL ml	URINE VOL ON DECALSO COLUMN ml	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL UNKNOWN ml	BIO-ASSAY HISTAMINE ST SOLN ml	HOURLY URINARY HISTAMINE μg	VOL GASTRIC JUICE SECRETED ml/hr	mEq HCl per hr
105 2	CONTROL HOUR	100	50	10	0·4	0·08	0·4		
	HOUR 1	35	35	30	0·06	0·04	1·99		
	HOUR 2	77	35·5	10	0·06	0·04	3·31		
	HOUR 3	32	32	10	0·04	0·04	1·0		
	HOUR 4	86	43	10	0·04	0·04	2·06		
	HOUR 5	65	32·5	10	0·12	0·04	0·67		
	HOUR 6	17	17	5	0·1	0·04	0·2		
	HOUR 7	114	50	5	0·12	0·04	0·38		
	HOUR 8	317	47·5	5	0·18	0·02	0·35		
	HOUR 9	157	35	5	0·1	0·02	0·45		
	HOUR 10								

HISTAMINE STANDARD — 0·1 μg./ml.

COMMENTARY ON RESULTS.

The results obtained in these experiments demonstrate that free histamine still appears in the urine after a meat meal is given to totally gastrectomised dogs. Since no similar experiments had been performed in these dogs prior to operation no quantitative comparisons of the amounts present in the urine before and after gastrectomy could be made. However, it appeared permissible to contrast the general pattern of histamine excretion obtained in these tests with those obtained in the previous chapter in dogs with stomachs. (Fig. 18 illustrates the mean of the two groups of tests).

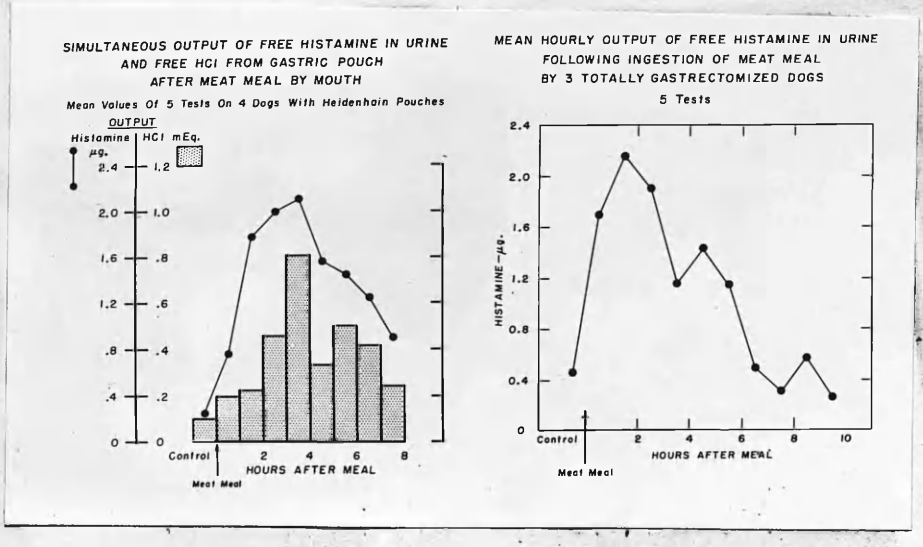


FIG. 18.

The means of the two groups of tests show certain differences. After total gastrectomy, when the meal passes directly into the small bowel unchecked by the pyloric sphincter, the rise in free histamine occurs more precipitiously, the peak being reached by the second hour. In contrast, dogs with intact stomachs have only a very moderate rise in the first hour and a high plateau occurs between the 3rd and 6th hours. After gastrectomy, the peak is not maintained and control levels are reached several hours sooner than occurs in the non-gastrectomised animal. It is not possible to say from these tests that the lower levels of urinary histamine seen between the 4th and 8th hours are related to the absence of gastric mucosa. It must be remembered that intestinal hurry is a marked feature of the post-gastrectomy state (Ivy, 1940: Emery, 1935: McCorkle and Harper, 1954). In this study the alimentation time of these animals after total gastrectomy was reduced to between 20 and 40% of that found pre-operatively, as measured by the carmine red test. Thus, if absorption from the small bowel played some part in the production of increased urinary histamine, it may have been affected by the removal of the stomach. However, these tests show clearly that considerable quantities of free histamine still appear in the urine after a meat meal when the gastric mucosa is completely absent.

CONCLUSIONS.

(1) The rise in free histamine in the urine after /

/after a meal of meat by mouth still occurs after total gastrectomy. This points against the gastric mucosa being the only source of this histamine in the urine.

(2) When the meat meal enters the small bowel directly the histamine in the urine rises more rapidly and remains elevated for a shorter period in the presence of a reduced alimentation time.

(3) Since no pre-operative data were available on these dogs, no quantitative comparisons of the effect of the operation on free histamine output were possible. That one or other phase of acid secretion contributes significant quantities of free histamine to the urine is not excluded by these experiments. This latter possibility was therefore studied further.

CHAPTER VI.

OUTPUT OF URINARY HISTAMINE DURING PSYCHIC OR NERVOUS GASTRIC SECRETION.

We have seen that the output of histamine in the urine after a meat meal was not abolished by total gastrectomy. In this and the following two chapters an attempt was made to isolate the psychic, gastric and intestinal phases of acid secretion to determine if any one of these components augmented the urinary histamine, and to correlate any such rise with the acid response produced. Although we are mainly concerned with the effects of a meal of meat on denervated gastric pouches, for completeness we required to know the effect of nervous gastric secretion on the urinary histamine. This chapter records such a study.

OBJECT OF EXPERIMENTS.

To measure the output of free histamine in the urine of dogs before and during acid secretion induced by vagal stimulation.

MATERIAL AND METHODS.

To study the nervous phase of acid secretion, an animal preparation, with intact vagal nerve supply to the gastric mucosa was required. /

/required.

The procedure of simple gastrostomy was carried out in three dogs by inserting a vitallium cannula in the most dependent part of the main stomach, without interrupting its blood or nerve supply. The cannula contained a plug which could readily be removed for the tests, to prevent loss of secretions.

VAGAL STIMULATION.

This was produced by inducing hypoglycaemia with a dose of intravenous insulin (2 units/kg. body weight). Blood sugar estimations were carried out to check that the blood sugar had fallen below 50 mg. per cent.

EXPERIMENTS.

After a 48 hour fast the dogs were catheterised and urine output maintained as previously described. Tests consisted of hourly collections of urine and acid for a control period of 2-3 hours. Following the insulin induced hypoglycaemia, collections were continued for a further 3-4 hours. Suction was applied to the stomach through the cannula to ensure that all the gastric juice secreted was removed. Acid output and urinary histamine were estimated as previously described./

/described.

RESULTS. Six tests in three dogs are recorded below.

DOG 628.

Fig.19.

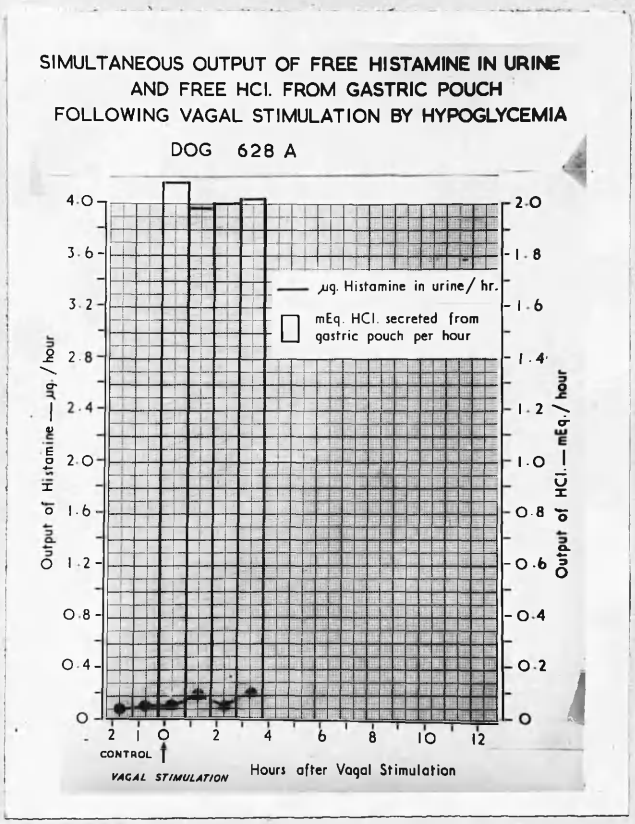
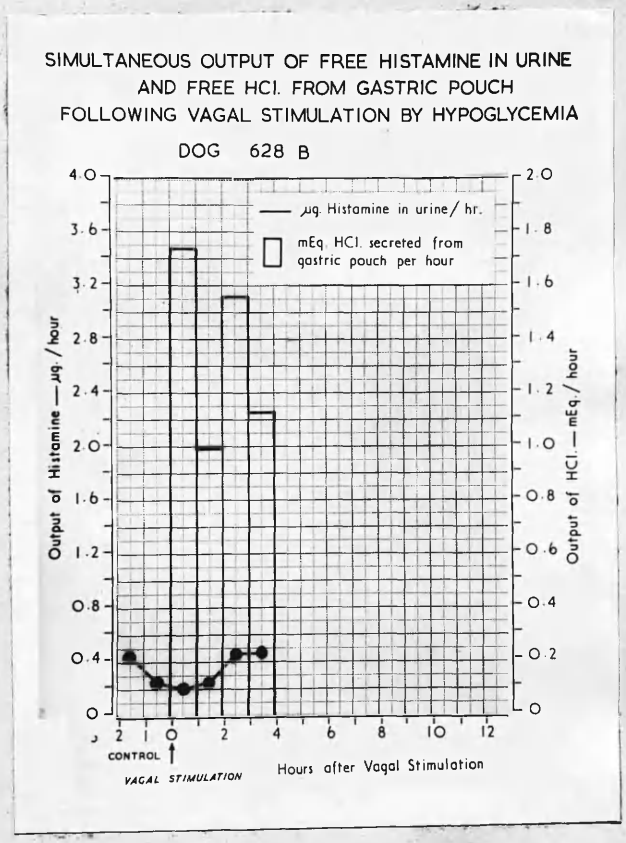


Fig.20.



EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE μg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
6 2 8 A	CONTROL HOUR 1	36	36	10	0·4	0·04		4·4	0
	CONTROL HOUR 2	36	38	"	"	0·05		2·5	0
	HOUR 1	46	46	"	"	0·05		44·0	2·5
	HOUR 2	102	50	"	"	0·04		26·5	1·98
	HOUR 3	182	50	"	"	0·03		25·0	2·05
	HOUR 4	164	50	"	"	0·03		22·0	2·13
	HOUR 5	HISTAMINE STANDARD — 0·1 μg. / ml.							
	HOUR 6								
	HOUR 7								
	HOUR 8								
	HOUR 9								

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE μg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
6 2 8 B	CONTROL HOUR 1	28	28	10	0·36	0·16	0·44	4·0	0·0
	CONTROL HOUR 2	32	32	"	0·4	0·1	0·25	2·0	0·0
	HOUR 1	46	46	"	0·4	0·08	0·2	23·0	1·74
	HOUR 2	40	40	"	0·4	0·1	0·25	15·0	0·99
	HOUR 3	186	50	"	0·4	0·1	0·46	17·0	1·56
	HOUR 4						0·46	18·0	1·13
	HOUR 5	HISTAMINE STANDARD — 0·1 μg. / ml.							
	HOUR 6								
	HOUR 7								
	HOUR 8								
	HOUR 9								

Fig.21 DOG 628.

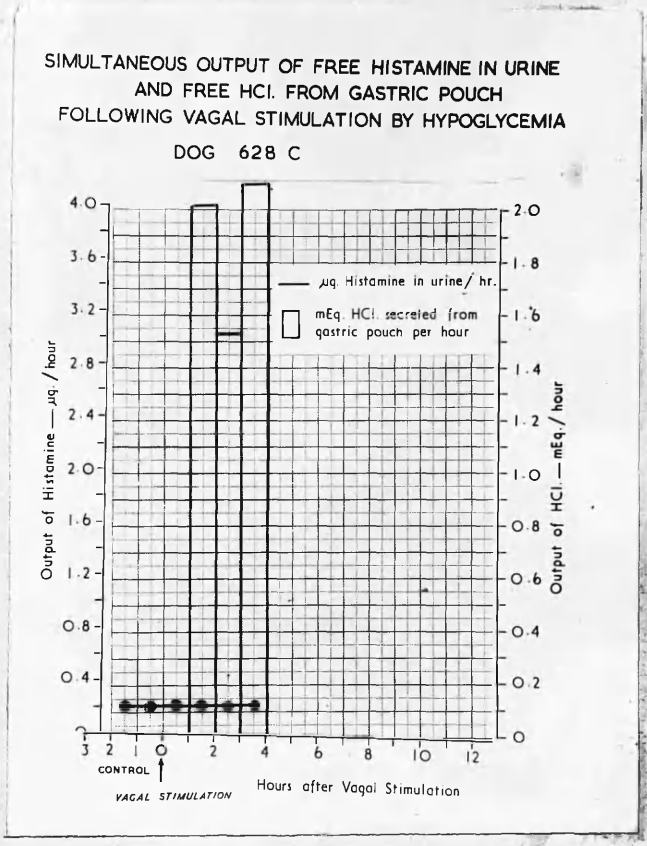
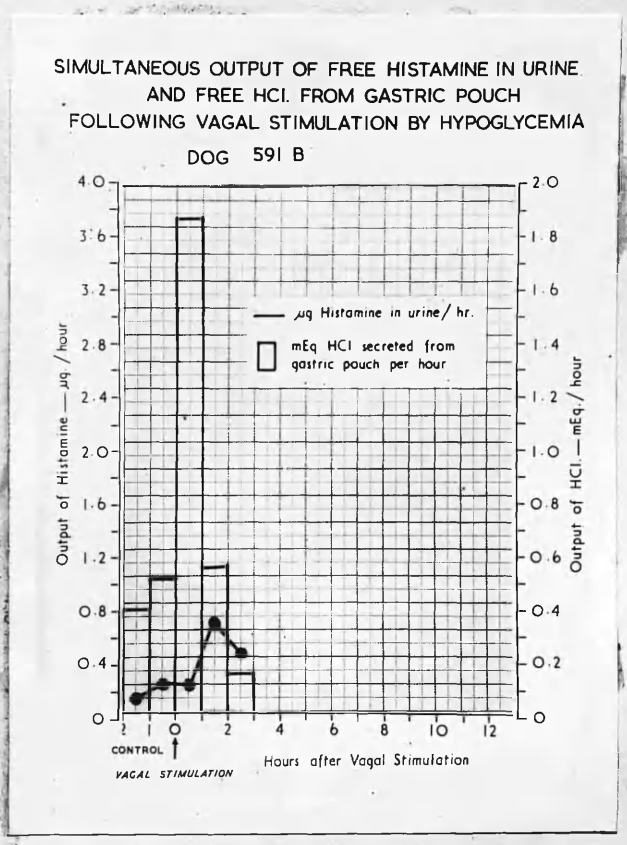


Fig.22. DOG 591.



EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE μg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR 1	21	21	10	0·4	0·06	0·15	0·6	0·0
6	CONTROL HOUR 2	23	23	"	0·4	0·06	0·15	3·0	0·0
2	CONTROL HOUR 3	29	29	"	0·4	0·06	0·15	5·0	0·0
8	HOUR 1	50	50	"	0·4	0·06	0·15	38·0	2·2
C.	HOUR 2	31	31	"	0·4	0·06	0·15	15·0	1·52
	HOUR 3	39	39	"	0·36	0·06	0·16	25·0	2·73
	HOUR 4			HISTAMINE STANDARD — 0·1 μg. / ml.					
	HOUR 5								
	HOUR 6								
	HOUR 7								
	HOUR 8								

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE μg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR 1	50	50	10	0·4	0·02	0·1	7·0	0·38
5	CONTROL HOUR 2	40	40	"	0·29	0·06	0·2	11·0	0·49
9	HOUR 1	48	48	"	0·28	0·06	0·21	24·5	1·81
1	HOUR 2	120	50	"	0·28	0·08	0·67	12·8	0·53
	HOUR 3	84	50	"	0·32	0·08	0·42	8·0	0·14
B.	HOUR 4								
	HOUR 5			HISTAMINE STANDARD — 0·1 μg. / ml.					
	HOUR 6								
	HOUR 7								
	HOUR 8								
	HOUR 9								

Fig.23.DOG 472.

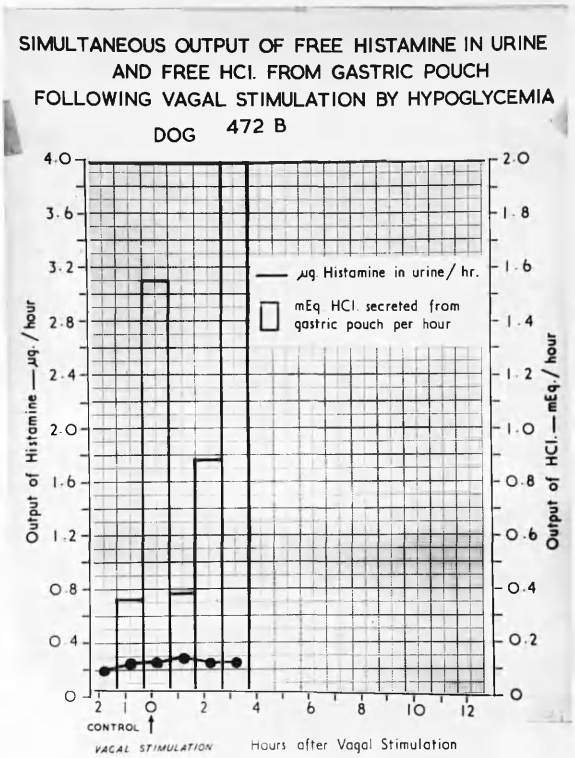
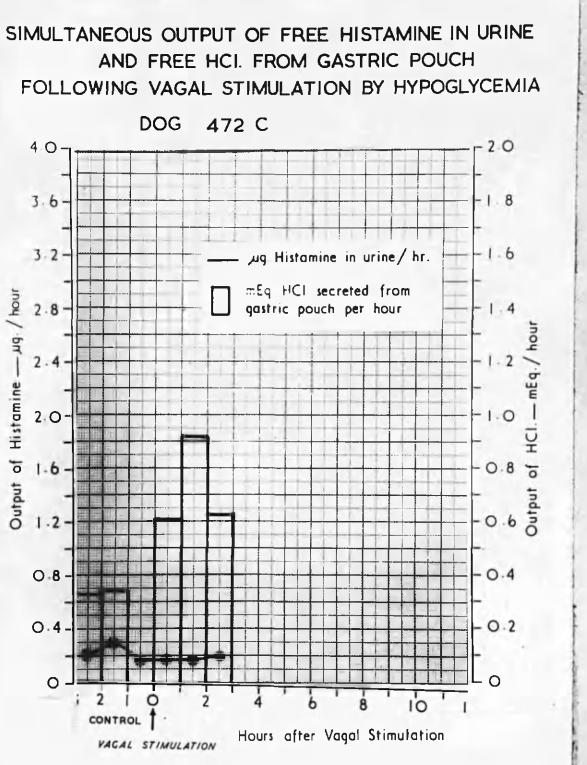


Fig.24. DOG 472.



EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE μg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
472B	CONTROL HOUR 1	16	16	10	0·4	0·08	0·2	10	0·0
	CONTROL HOUR 2	34	34	-	0·4	0·1	0·25	19	0·36
	HOUR 1	44	44	-	0·4	0·1	0·25	50	1·55
	HOUR 2	67	67	-	0·44	0·1	0·29	15·5	0·39
	HOUR 3	200	50	-	0·4	0·05	0·25	24·0	0·88
	HOUR 4							26·0	2·0
	HOUR 5			HISTAMINE STANDARD — 0·1 μg. / ml.					
	HOUR 6								
	HOUR 7								
	HOUR 8								
	HOUR 9								

EXP.	SPECIMEN	TOTAL URINE VOL ml	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST SOLN. ml	HOURLY URINARY HISTAMINE μg.	VOL GASTRIC JUICE SECRETED ml./hr	mEq HCl per hr.
472C	CONTROL HOUR 1	26	26	10	0·4	0·1	0·25	13·0	0·34
	CONTROL HOUR 2	44	44	-	0·4	0·12	0·3	17·0	0·34
	CONTROL HOUR 3	41	41	-	0·4	0·07	0·175	46·0	0·0
	HOUR 1	50	50	-	0·4	0·07	0·175	34·0	0·61
	HOUR 2	47	47	-	0·4	0·07	0·175	27·0	0·92
	HOUR 3	32	32	-	0·4	0·04	0·1	24·0	0·62
	HOUR 4			HISTAMINE STANDARD — 0·1 μg. / ml.					
	HOUR 5								
	HOUR 6								
	HOUR 7								
	HOUR 8								

COMMENTARY ON RESULTS.

One of the three dogs with gastrostomies used for these studies continued to have a considerable output of acid from the stomach even after 48 hours of fasting. In the remaining two animals, however, a 48 hour fast reduced the output of acid of the entire stomach to negligible proportions and five satisfactory tests were carried out in these two animals. The mean of these 5 tests in 2 dogs is shown in Fig. 25.

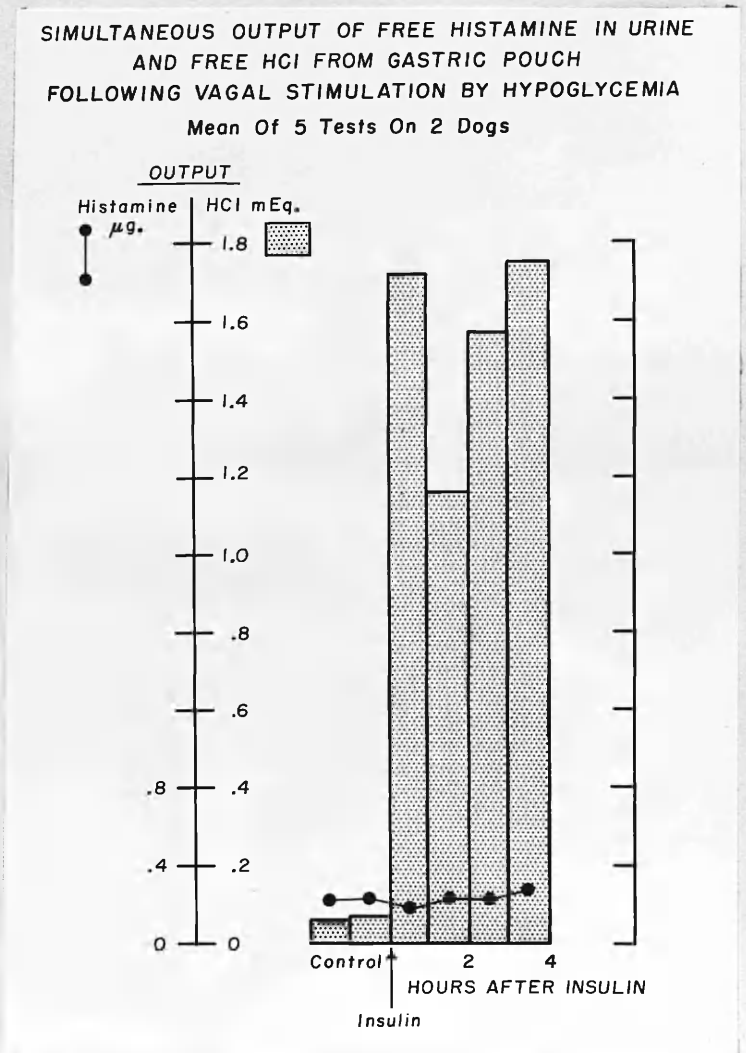


FIG. 25

During the period of hypoglycaemia, the output of acid from the stomach of these dogs usually reached levels of 1.5 Meq. per hour, and this output was sustained for the next 3-4 hours. During this period of increased acid output from the stomach, the histamine in the urine was unchanged, the low control levels being continued throughout the entire period of the tests (Fig. 19 - 23).

Even in the unsatisfactory third dog with high control levels of acid secretion, hypoglycaemia greatly augmented the acid secretion without raising the urinary histamine (Fig. 24).

CONCLUSIONS.

Vagal stimulation, produced by insulin induced hypoglycaemia, in dogs with simple gastrostomies, greatly augments acid secretion without affecting the output of free histamine in the urine.

CHAPTER VII.

OUTPUT OF FREE URINARY HISTAMINE DURING THE GASTRIC HORMONAL PHASE OF ACID SECRETION.

It has been seen that vagal stimulation can produce copious acid secretion from the gastric mucosa without increasing the output of free histamine in the urine. Of much greater interest was a study of the hormonal phase of acid secretion which has been attributed to the release of gastrin from the antrum.

A meat meal increases the free histamine in the urine and meat in contact with the antrum is a potent stimulator of acid secretion from a denervated gastric pouch. Does this antral hormonal phase of gastric secretion contribute any part to the increased output of free histamine in the urine after a meal? The studies reported below attempted to answer this question.

OBJECTIVE OF EXPERIMENTS.

To observe the effect on urinary histamine and acid secretion from denervated pouches of dogs, before and after the introduction of meat directly into the isolated stomach.

MATERIAL AND METHODS.

(a) ANIMAL PREPARATION.

Four dogs with denervated Heidenhain pouches had Thomas Cannulae inserted in their upper jejunum 4" beyond /

/beyond its commencement from the duodenum. These cannulae (Fig.26).

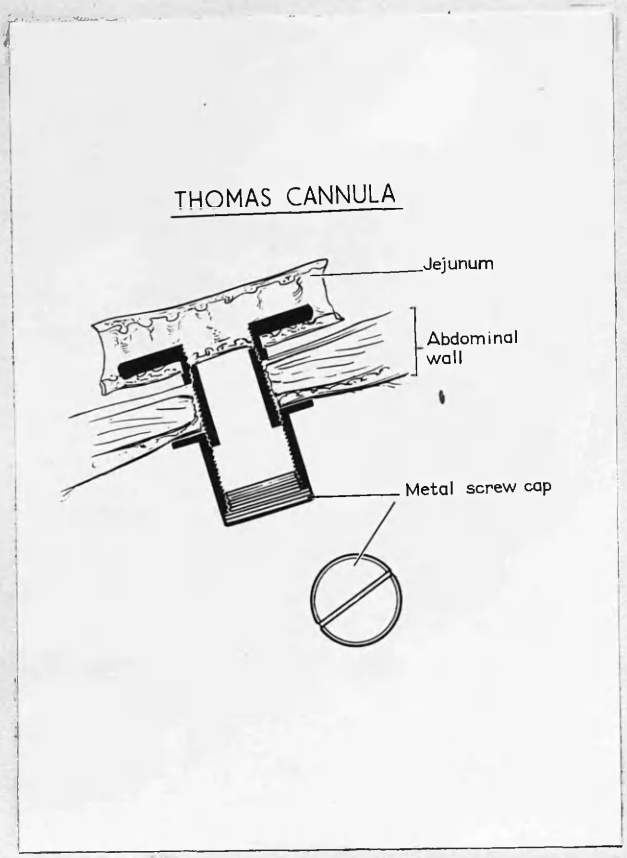


FIG. 26

are fitted with a screw cap which on removal permitted the installation of food directly into the small bowel. At a later operation, the pyloric end of the stomach was detached from the duodenum and both ends carefully inturned. A second vitallium cannula was then placed in the isolated stomach. Unlike the previous gastrostomy dogs this cannula was not plugged, thus preventing the accumulation of secretions with overflow into the bronchii. This animal preparation is represented diagrammatically in Fig. 27.

/in Fig. 27.

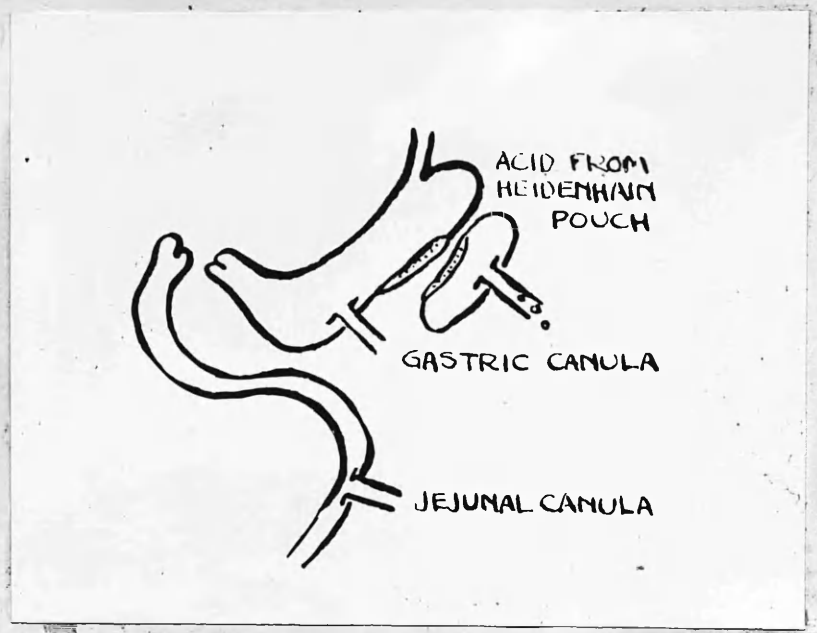


FIG. 27

Following operation these dogs were maintained by daily intravenous infusions and by jejunal feeding through the Thomas cannulae. These latter feeds were mainly of milk, a poor stimulator of acid secretion, to reduce the loss of chloride ions from the cannula in the main stomach.

TESTS.

Following the usual fast the dogs were catheterised and I.V. infusions commenced to maintain urine output. A meal of cooked horse meat (170 gms.) from the same source /

/source as that previously used, was homogenised in a Waring Blender and introduced into the body of the stomach through the gastric cannula. The urinary histamine and acid output from the gastric pouch were then measured at hourly intervals for the next 4 hours by the usual methods. The meal was then removed from the stomach and inserted into the upper jejunum through the Thomas cannula. Hourly collections of urine and acid were then continued for a further 5 hours.

CONTROL TESTS.

Each dog had hourly estimations of acid and urinary histamine following I.V. infusions of histamine at fixed dose rates. The purpose of these tests will be described later.

RESULTS.

Eight tests were carried out on four dogs. Graphs illustrating the results of each test will be presented on pages 56 and 57. The tables from which the graphs were drawn are included in the following two pages (p. 58 and 58a).

Fig. 28

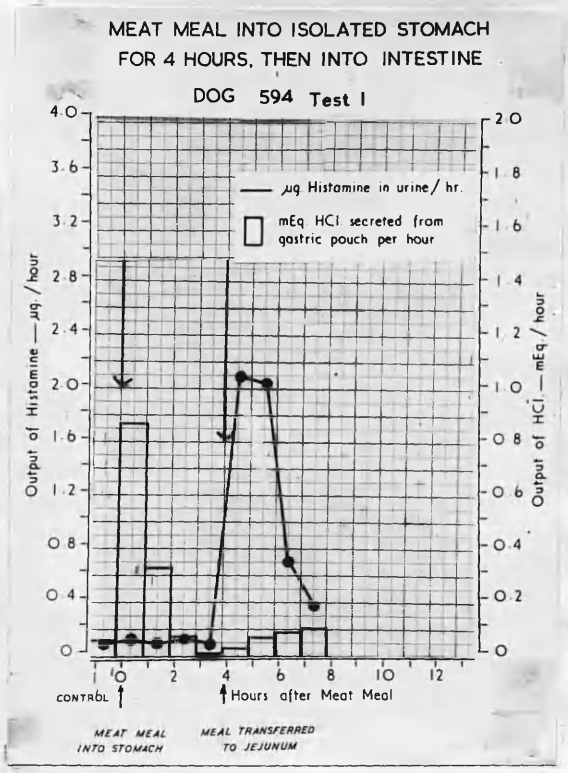


Fig. 29

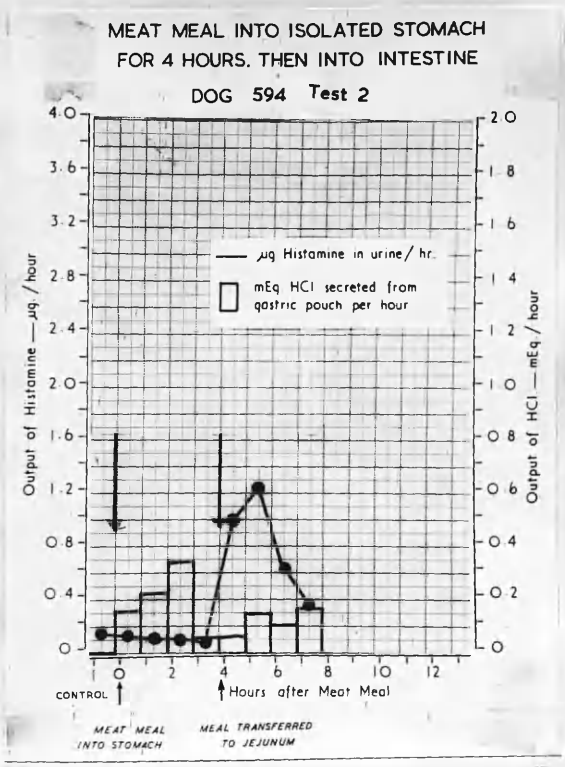


Fig. 30

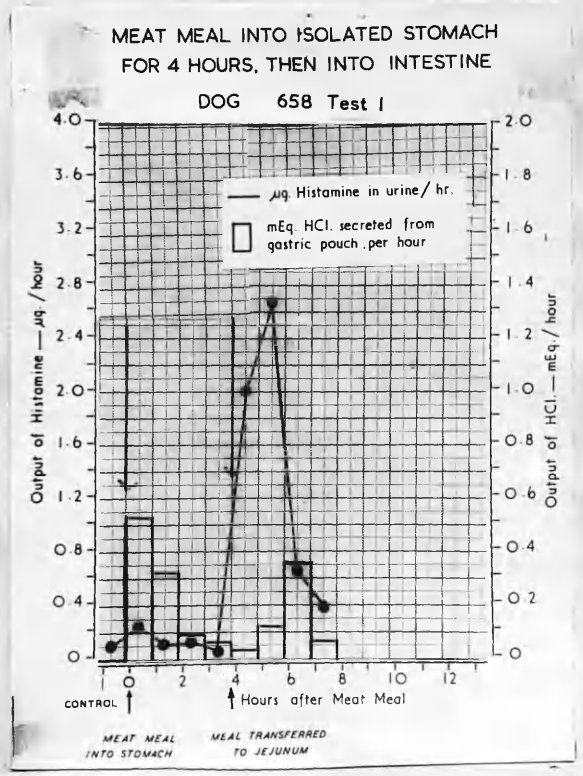


Fig. 31

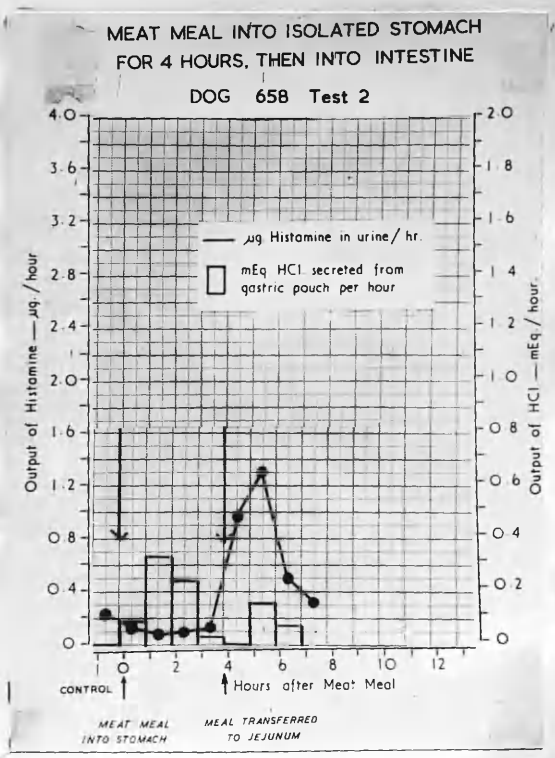


Fig. 32

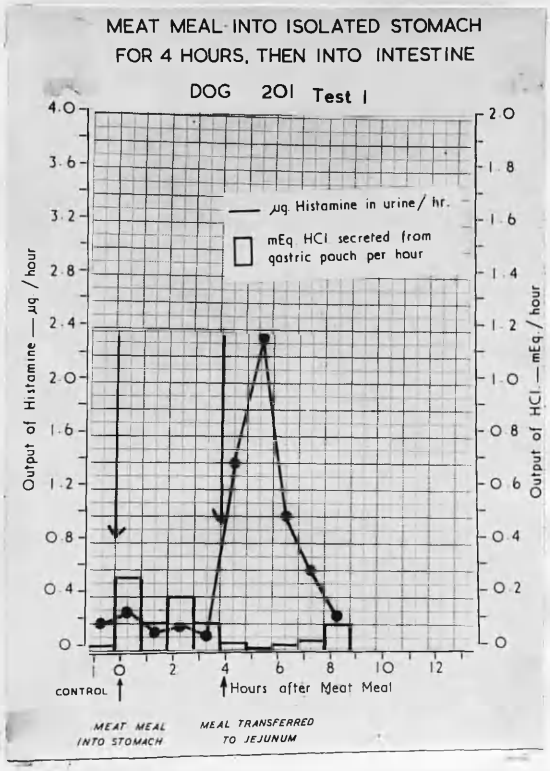


Fig. 33

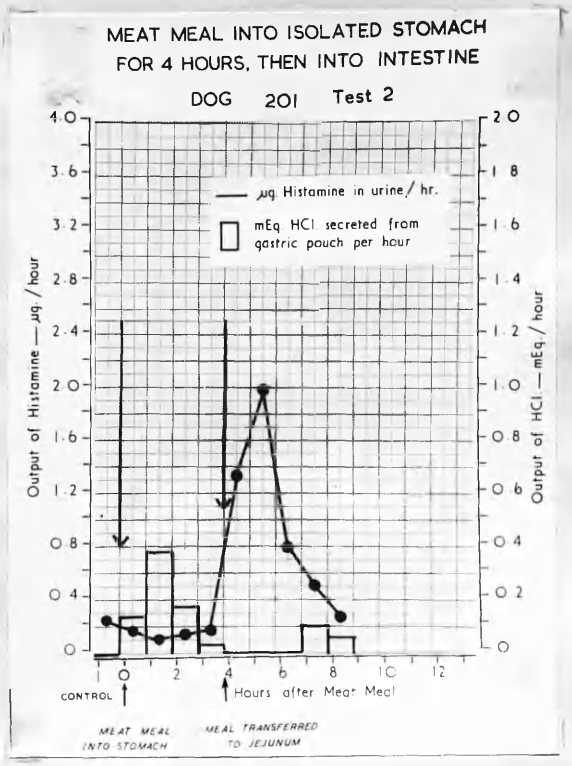


Fig. 34

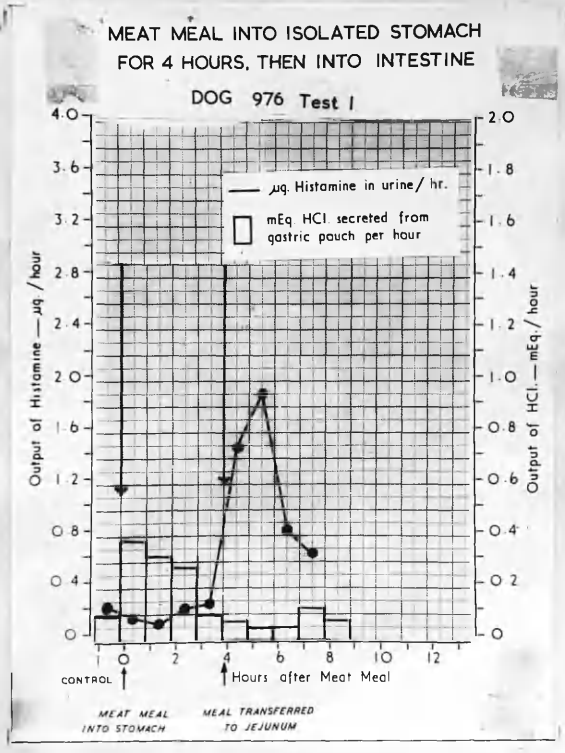
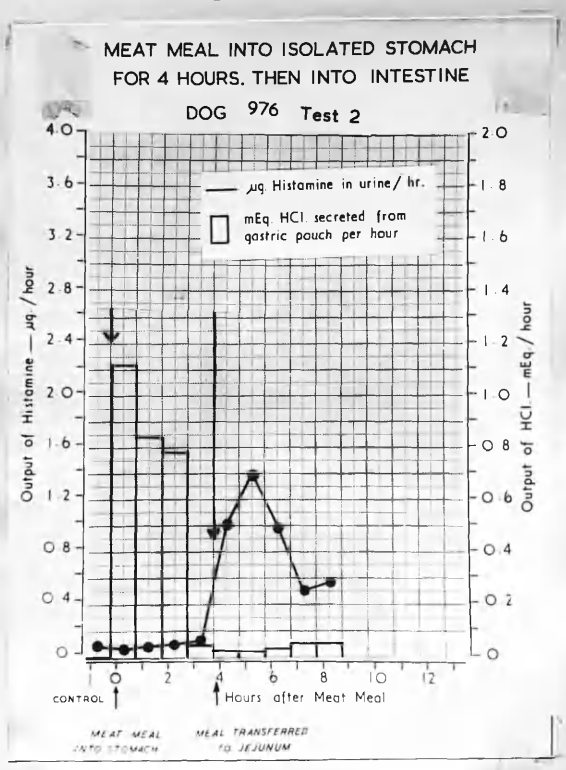


Fig. 35



Dog 594

Data from which figures 28 and 29 were drawn.

Test 1

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL GASTRIC JUICE SECRETED ml./hr.	mEq HCl. per hr.
	CONTROL HOUR	48	48	10	0·4	0·04	0·1	1·2	0·06
5	HOUR 1	129	50	"	0·4	0·02	0·13	10·5	0·87
9	HOUR 2	50	50	"	0·4	0·04	0·1	4·5	0·33
4	HOUR 3	80	50	"	0·4	0·03	0·12	1·1	0·07
	HOUR 4	55	50	"	0·4	0·03	0·075	0·9	0·01
TEST	HOUR 5	66	50	"	0·05	0·08	2·1	1·3	0·03
	HOUR 6	117	50	"	0·09	0·08	2·05	1·6	0·07
1.	HOUR 7	80	50	"	0·18	0·08	0·7	1·3	0·09
	HOUR 8	50	50	"	0·22	0·08	0·36	1·5	0·1
	HOUR 9								
	HOUR 10								

HISTAMINE STANDARD — 0·1 µg. / ml.

Test 2

EXP.	SPECIMEN	TOTAL URINE VOL ml	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq HCl. per hr.
	CONTROL HOUR	34	34	10	0·4	0·06	0·15	0·9	0·0
5	HOUR 1	32	32	"	0·4	0·05	0·13	3·3	0·16
9	HOUR 2	44	44	"	0·4	0·04	0·1	2·8	0·23
	HOUR 3	50	50	"	0·4	0·04	0·1	4·0	0·34
4	HOUR 4	80	50	"	0·4	0·02	0·08	1·0	0·06
	HOUR 5	97	50	"	0·16	0·08	1·0	1·2	0·06
TEST	HOUR 6	108	50	"	0·06	0·04	1·23	2·1	0·15
	HOUR 7	68	50	"	0·13	0·06	0·62	1·5	0·11
2.	HOUR 8	50	50	"	0·16	0·06	0·37	2·0	0·16
	HOUR 9								
	HOUR 10								

HISTAMINE STANDARD — 0·1 µg. / ml.

Dog 658

Date from which figures 30 and 31 were drawn.

Test 1

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL GASTRIC JUICE SECRETED ml./hr.	mEq HCl. per hr.
	CONTROL HOUR	34	34	10	0·4	0·04	0·1	0·0	0·0
6	HOUR 1	127	50	"	0·4	0·04	0·25	8·2	0·5
5	HOUR 2	75	50	"	0·4	0·03	0·11	4·3	0·32
8	HOUR 3	84	50	"	0·4	0·03	0·12	1·8	0·09
	HOUR 4	50	50	"	0·4	0·02	0·05	1·9	0·06
TEST	HOUR 5	50	50	"	0·05	0·1	2·0	1·7	0·03
	HOUR 6	122	50	"	0·09	0·1	2·68	3·0	0·12
1.	HOUR 7	100	50	"	0·24	0·08	0·66	4·0	0·36
	HOUR 8	53	50	"	0·2	0·06	0·37	1·0	0·6
	HOUR 9								
	HOUR 10								

HISTAMINE STANDARD — 0·1 µg. / ml.

Test 2

EXP.	SPECIMEN	TOTAL URINE VOL. ml	URINE VOL ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL GASTRIC JUICE SECRETED ml./hr.	mEq HCl. per hr.
6	CONTROL HOUR	34	34	10	0·36	0·08	0·22	0·0	0·0
5	HOUR 1	85	50	"	0·4	0·03	0·129	2·2	0·09
8	HOUR 2	50	50	"	0·4	0·03	0·075	4·5	0·35
	HOUR 3	98	50	"	0·4	0·02	0·098	2·7	0·24
7	HOUR 4	61	50	"	0·4	0·04	0·12	0·8	0·03
TEST	HOUR 5	73	50	"	0·12	0·08	0·96	0·4	0·0
	HOUR 6	76	50	"	0·07	0·06	1·3	3·2	0·16
2.	HOUR 7	50	50	"	0·12	0·06	0·5	1·0	0·07
	HOUR 8	50	50	"	0·18	0·06	0·33	0·0	0·0
	HOUR 9								
	HOUR 10								

HISTAMINE STANDARD — 0·1 µg. / ml

Dog 201

Date from which figures 32 and 33 were drawn.

Test 1

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
201 TEST 1.	CONTROL HOUR	50	50	10	0·2	0·4	0·2	1·0	0·17
	HOUR 1	105	50	"	0·28	0·04	0·29	4·6	0·27
	HOUR 2	67	50	"	0·4	0·04	0·13	1·9	0·12
	HOUR 3	122	50	"	0·4	0·03	0·18	3·2	0·2
	HOUR 4	82	50	"	0·07	0·03	0·12	2·8	0·11
	HOUR 5	50	50	"	0·08	0·1	1·4	1·0	0·03
	HOUR 6	118	50	"	0·12	0·08	2·36	0·3	0·01
	HOUR 7	124	50	"	0·15	0·05	1·04	1·0	0·02
	HOUR 8	121	50	"	0·4	0·04	0·6	2·6	0·04
	HOUR 9	48	48	"		0·1	0·25	2·6	0·1
	HOUR 10								

HISTAMINE STANDARD — 0·1 µg. / ml.

Test 2

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
201 TEST 2.	CONTROL HOUR	40	40	10	0·4	0·1	0·25	0·8	0·0
	HOUR 1	45	45	"	0·36	0·06	0·17	3·0	0·14
	HOUR 2	50	50	"	0·4	0·04	0·1	4·4	0·38
	HOUR 3	79	50	"	0·4	0·04	0·158	2·0	0·17
	HOUR 4	124	50	"	0·4	0·03	0·86	1·3	0·03
	HOUR 5	50	50	"	0·06	0·08	1·33	0·5	0·0
	HOUR 6	124	50	"	0·1	0·08	1·99	0·5	0·0
	HOUR 7	75	50	"	0·15	0·08	0·79	0·5	0·0
	HOUR 8	35	35	"	0·16	0·08	0·5	2·8	0·1
	HOUR 9	32	32	"	0·28	0·08	0·27	1·7	0·06
	HOUR 10								

HISTAMINE STANDARD — 0·1 µg. / ml.

Dog 976

Date from which figures 34 and 35 were drawn.

Test 1

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
976 TEST 1.	CONTROL HOUR	50	50	10	0·16	0·04	0·25	3·2	0·09
	HOUR 1	122	50	"	0·3	0·02	0·17	8·0	0·38
	HOUR 2	72	50	"	0·35	0·03	0·12	4·2	0·32
	HOUR 3	119	50	"	0·3	0·03	0·24	5·0	0·28
	HOUR 4	103	50	"	0·3	0·04	0·27	3·2	0·09
	HOUR 5	148	50	"	0·8	0·04	1·48	2·3	0·07
	HOUR 6	119	50	"	0·1	0·08	1·9	1·6	0·05
	HOUR 7	84	50	"	0·16	0·08	0·84	1·6	0·05
	HOUR 8	83	50	"	0·2	0·08	0·66	2·3	0·12
	HOUR 9	40	40	"	0·38	0·1	0·26	1·6	0·07
	HOUR 10								

HISTAMINE STANDARD — 0·1 µg. / ml.

Test 2

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
976 TEST 2.	CONTROL HOUR	40	40	10	0·4	0·04	0·1	0·6	0·00
	HOUR 1	72	50	"	0·4	0·02	0·07	10·2	1·12
	HOUR 2	63	50	"	0·4	0·03	0·09	8·0	0·84
	HOUR 3	50	50	"	0·4	0·04	0·1	8·4	0·78
	HOUR 4	85	50	"	0·4	0·03	0·13	1·2	0·04
	HOUR 5	68	50	"	0·08	0·06	1·02	1·0	0·02
	HOUR 6	153	50	"	0·12	0·05	1·29	1·0	0·02
	HOUR 7	97	50	"	0·16	0·08	0·97	1·7	0·03
	HOUR 8	31	31	"	0·16	0·08	0·3	2·2	0·05
	HOUR 9	39	39	"	0·14	0·08	0·57	2·1	0·05
	HOUR 10								

HISTAMINE STANDARD — 0·1 µg. / ml.

COMMENTARY ON RESULTS.

From the results it will be seen that two tests were carried out in each dog. The results were uniform. The mean of the 8 tests in 4 dogs is shown in Fig. 36.

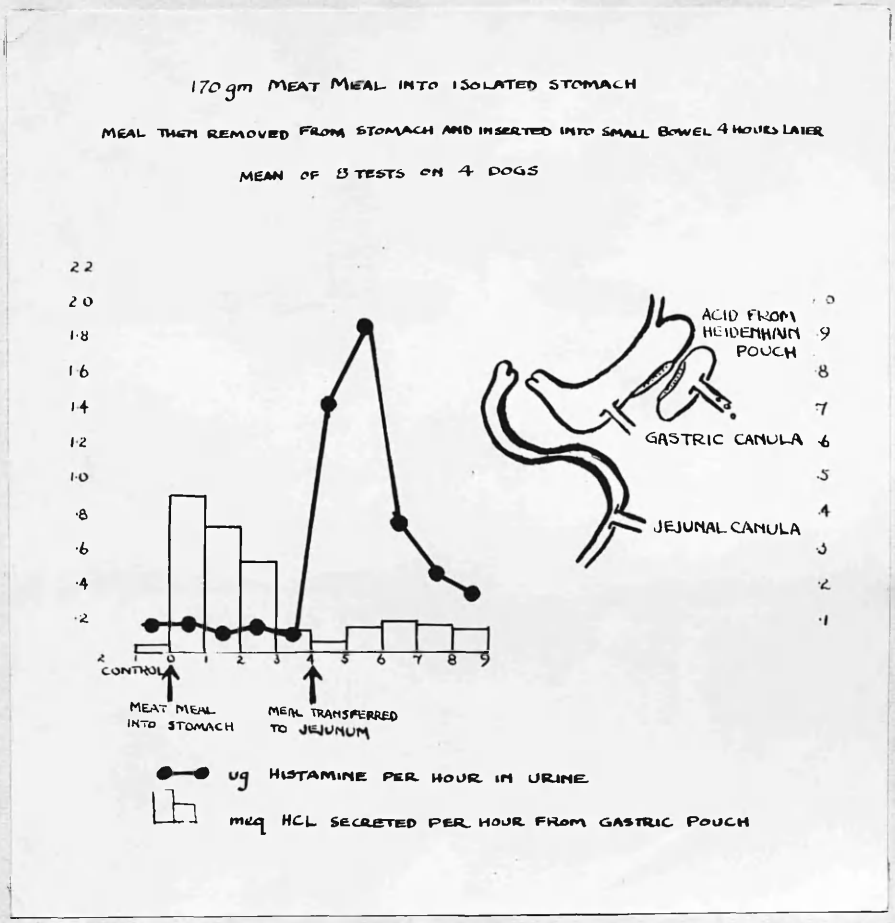


FIG. 36.

While the meat meal resided in the stomach a good acid response was obtained in the denervated Heidenhain pouch, but there was no rise in the free histamine in the urine above control values during this period. When the meal was removed /

/removed from the stomach and inserted into the jejunum, a rapid rise in urinary histamine promptly occurred, the peak being reached in the second hour. The histamine level then fell rapidly to control levels. With the meat in the intestine the acid response, though small, was maximal only after a time lag of 2-4 hours. Figs. 28 - 35.

Thus the meat in the isolated stomach in contact with the antrum caused a substance (referred to here as Gastrin) to enter the blood stream and stimulate the denervated gastric pouch. This hormonal stimulation of acid secretion was not associated with any detectable rise in the free histamine in the urine when the meat was in the isolated stomach.

Would a slight rise in free circulating histamine sufficient to produce the modest stimulation of acid obtained from the pouches of these dogs be reflected in an increase of urinary histamine, detectable by the method used? To test this each dog had a series of I.V. infusions of histamine at fixed rates, the dose being reduced until the gastric pouch gave a response roughly equivalent to that obtained when the meat was in contact with the antrum. In each dog when the level of free circulating histamine was thus raised to give an equivalent acid response, considerable increases in the output of free histamine in the urine were detected. Two such control studies, placed beside the response to antral stimulation previously recorded, are illustrated in /

/in Figs. 37 and 38.

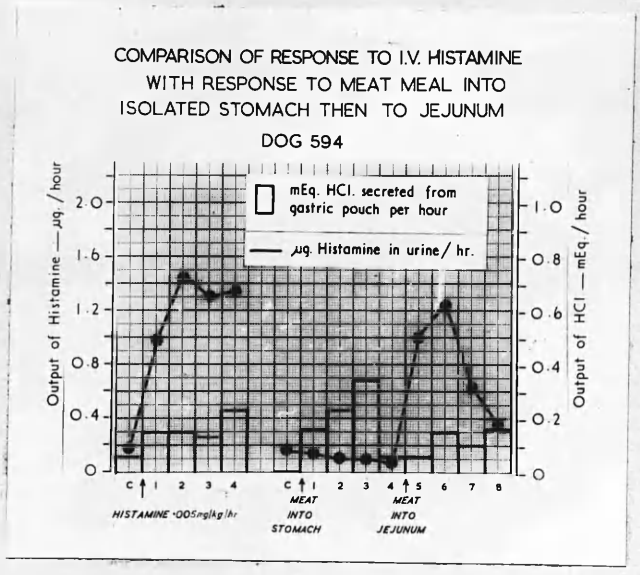


FIG. 37

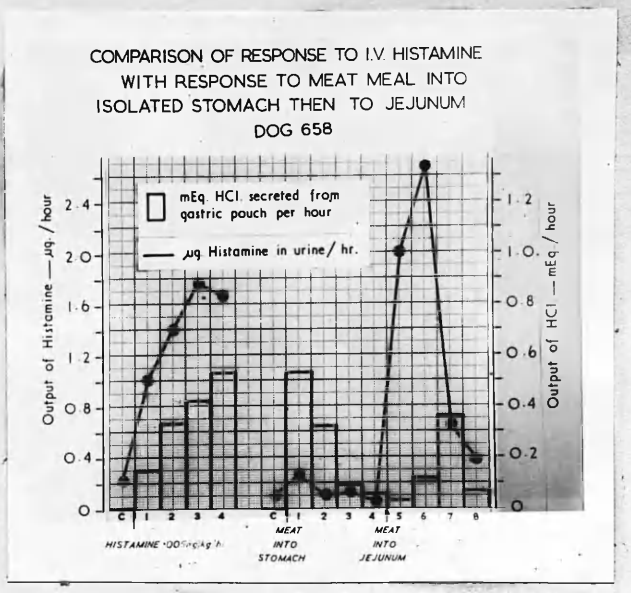


FIG. 38

One is forced to conclude that the substance entering the blood stream from the stomach wall which stimulated the denervated pouch did not produce its effect by raising the level of free histamine in the general circulation. Nor could it be a general histamine liberator releasing histamine from the tissues of the body, the subsequent rise in blood histamine stimulating acid secretion. It follows that gastrin is unlikely to be free histamine, nor can it be a general histamine liberator.

These experiments do not exclude the possibility that gastrin produces its effect by releasing histamine locally at the parietal cell. It may be said, however, that if gastrin does release histamine at the parietal cell, insufficient of it is absorbed back into the general circulation to be detected in the urine by the method used.

CONCLUSIONS.

(1) Meat in contact with the antrum, releases into the blood stream a substance, gastrin, which stimulates acid secretion from a denervated pouch without any rise in the urinary histamine being detected by the method used.

(2) When an equivalent acid response from each pouch is produced by an I.V. infusion of histamine, a considerable elevation of urinary histamine is detectable by the same method.

(3) These observations point against gastrin being /

/being histamine or a general histamine liberator. They do not exclude the possibility that gastrin releases histamine locally at the parietal cell.

(4) Meat only augments the urinary histamine after entering the small bowel. When meat is placed in the isolated stomach for several hours the output of histamine in the urine remains low. This same meat meal when removed from the stomach and inserted into the jejunum produces a rapid increase in free histamine in the urine.

CHAPTER VIII.

THE INTESTINAL PHASE OF ACID SECRETION.

In the experiments recorded in the previous chapter it was observed that meat, when placed in the isolated stomach, induced an acid response in a denervated gastric pouch without any elevation of urinary histamine. On the other hand, the same meat meal when placed in the small bowel, produced a rapid increase in urinary histamine excretion, usually associated with a very small acid response from the gastric pouch. The dogs, with interruption of alimentary tract continuity, tired easily and these observations on the effect of placing meat in the small bowel were commenced after the dogs had already been on the stands for 5 hours. Similar studies of the intestinal phase of acid secretion carried out in the fresh dog, prior to division of its duodeno-pyloric junction, are recorded in this chapter.

OBJECTIVE OF EXPERIMENTS.

To study the changes in urinary histamine and acid secretion from denervated gastric pouches of dogs, before and after introduction of a meal directly into the small bowel.

MATERIAL AND METHODS.

Female mongrel dogs of 9-15 kilo body weight /

/weight were used. Each dog had a denervated gastric pouch of Heidenhain type and a Thomas cannula placed in the upper jejunum 8" beyond the ligament of Treitz. Since a barium meal introduced into the jejunum at this point was invariably seen under fluoroscopy to regurgitate into the stomach, a further cannula was placed in the main stomach. Regurgitating meat would then be removed from contact with the antrum, by aspiration through this cannula. A diagrammatic representation of this animal preparation is shown in Fig. 38a

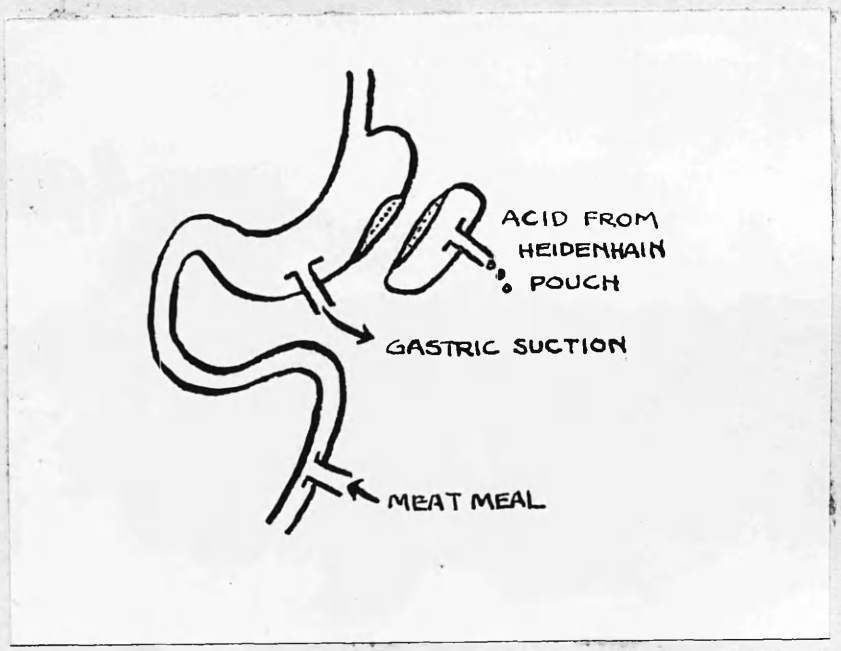


FIG. 38a

Experiments consisted as before of simultaneous hourly collections of acid and urine before and after the intestinal feed by the usual methods. /

/methods.

In approximately half of these tests a wide bore tube was passed into the main stomach through its cannula and continuous suction applied for 3 hours following introduction of the meal into the small bowel.

On most occasions, the intestinal feed consisted of 170 gms. of cooked horse meat which was homogenised in a Waring blender and introduced through the Thomas cannula into the jejunum over a 20 minute period.

Each dog had one intestinal feed of bread and milk whose total volume and calorific value equalled that of the meat meal. It was introduced in a similar manner through the Thomas cannula.

CONTROL STUDIES.

(1) INTRAVENOUS HISTAMINE TESTS.

Each dog had continuous infusions of histamine at different dose rates, acid output and urinary histamine being measured in the usual way.

(2) SUBCUTANEOUS HISTAMINE TEST.

The effect of introducing a meat meal into the jejunum, on the acid response to subcutaneous histamine, was measured in one dog.

The purpose of these tests will be discussed later.

RESULTS.

The results on the 4 dogs are illustrated in the following figures.

Dog 594

MEAT INTO SMALL BOWEL WITH GASTRIC SUCTION

Fig. 38 b

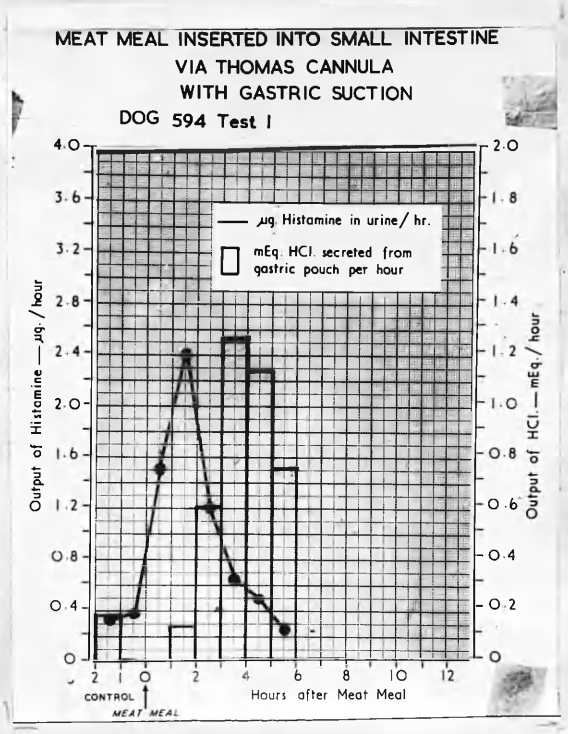
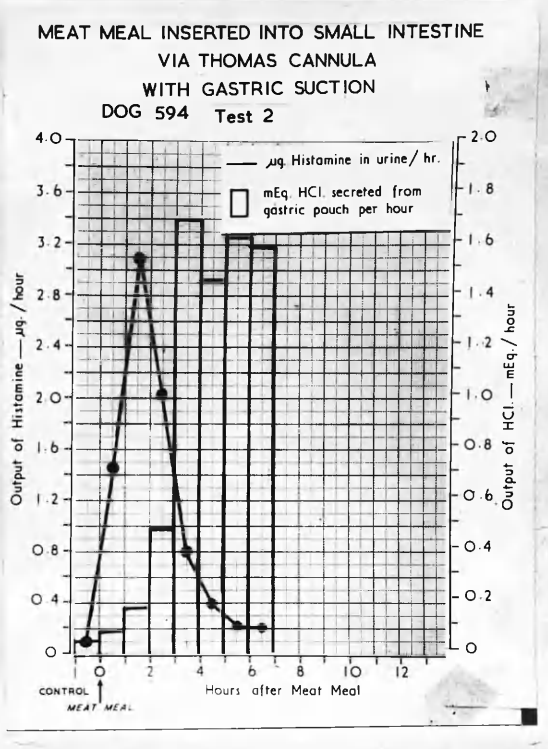


Fig. 39



MEAT INTO SMALL BOWEL WITHOUT GASTRIC SUCTION

Fig. 40

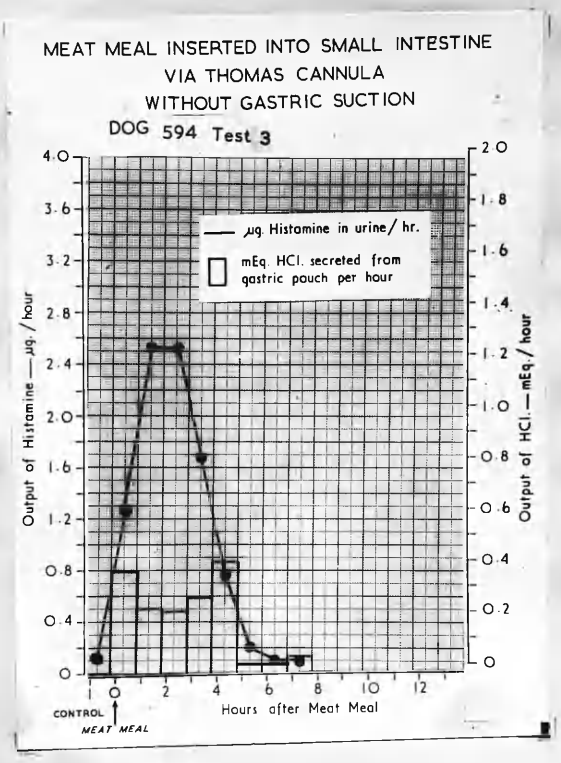
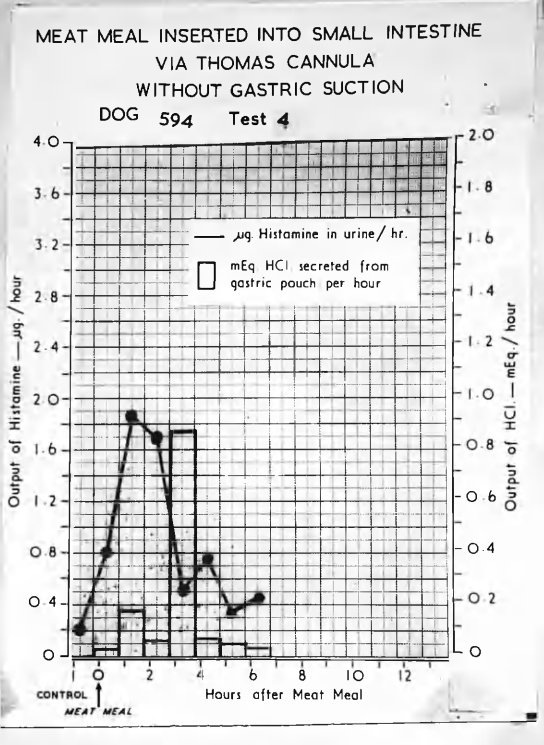


Fig. 41



Data from which the graphs for dog 594 were constructed (fig. 38b - 41).

Fig. 42.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl per hr.
5 9 4. TEST 1.	CONTROL HOUR 1	167	50	10	0·4	0·04	0·33	1·2	0·18
	CONTROL HOUR 2	198	50	"	0·4	0·04	0·39	4·5	0·01
	HOUR 1	75	50	"	0·12	0·12	1·5	0·8	0·01
	HOUR 2	50	50	"	0·05	0·12	2·4	0·9	0·14
	HOUR 3	54	54	"	0·10	0·12	1·2	2·5	0·61
	HOUR 4	52	52	"	0·18	0·12	0·66	6·5	1·27
	HOUR 5	48	48	"	0·2	0·1	0·5	10·5	1·14
	HOUR 6	31	31	"	0·22	0·06	0·27	9·5	0·76
	HOUR 7								
	HOUR 8								
	HOUR 9								

HISTAMINE STANDARD — 0·1 µg / ml.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl per hr.
5 9 4 TEST 2.	CONTROL HOUR	95	50	10	0·4	0·02	0·095	1·0	0·05
	HOUR 1	110	50	"	0·12	0·08	1·45	2·3	0·09
	HOUR 2	69	50	"	0·035	0·08	3·16	3·0	0·18
	HOUR 3	70	50	"	0·055	0·08	2·03	5·7	0·49
	HOUR 4	50	50	"	0·1	0·08	0·8	9·0	1·7
	HOUR 5	56	56	"	0·2	0·08	0·4	14·5	1·46
	HOUR 6	26	26	"	0·26	0·06	0·23	14·2	1·63
	HOUR 7	55	55	"	0·3	0·06	0·2	13·9	1·59
	HOUR 8								
	HOUR 9								
	HOUR 10								

HISTAMINE STANDARD — 0·1 µg / ml.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl per hr.
5 9 4 TEST 3.	CONTROL HOUR	26	26	10	0·4	0·04	0·1	1·2	0·0
	HOUR 1	30	30	-	0·08	0·1	1·25	4·9	0·39
	HOUR 2	115	50	-	0·09	0·1	2·53	2·7	0·25
	HOUR 3	63	50	-	0·05	0·1	2·52	3·0	0·25
	HOUR 4	67	50	-	0·08	0·1	1·57	3·0	0·29
	HOUR 5	77	50	-	0·12	0·06	0·76	4·8	0·43
	HOUR 6	30	30	-	0·2	0·04	0·2	1·4	0·04
	HOUR 7	47	47	-	0·4	0·04	0·1	1·2	0·04
	HOUR 8	29	29	-	0·4	0·04	0·1	0·7	0·02
	HOUR 9								
	HOUR 10								

HISTAMINE STANDARD — 0·1 µg / ml.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl per hr.
5 9 4 TEST 3.	CONTROL HOUR	30	30	10	0·4	0·08	0·2	1·8	0·0
	HOUR 1	39	39	-	0·25	0·2	0·8	1·3	0·0
	HOUR 2	250	50	-	0·32	0·12	1·87	2·3	0·1
	HOUR 3	94	50	-	0·22	0·2	1·69	1·0	0·0
	HOUR 4	50	50	-	0·1	0·2	0·5	7·8	0·8
	HOUR 5	31	31	-	0·24	0·18	0·75	0·7	0·0
	HOUR 6	47	47	-	0·24	0·08	0·33	1·0	0·0
	HOUR 7	24	24	-	0·18	0·08	0·44	0·7	0·0
	HOUR 8								
	HOUR 9								
	HOUR 10								

HISTAMINE STANDARD — 0·1 µg / ml.

Dog 658

MEAT INTO SMALL BOWEL WITH GASTRIC SUCTION

Fig. 43

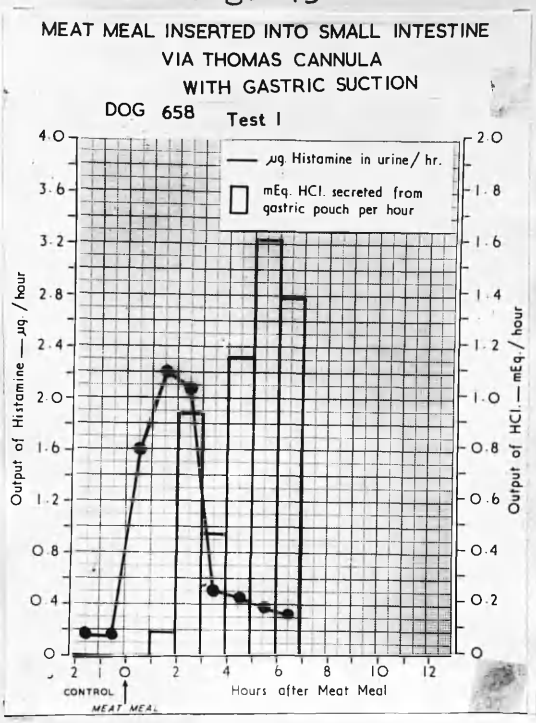
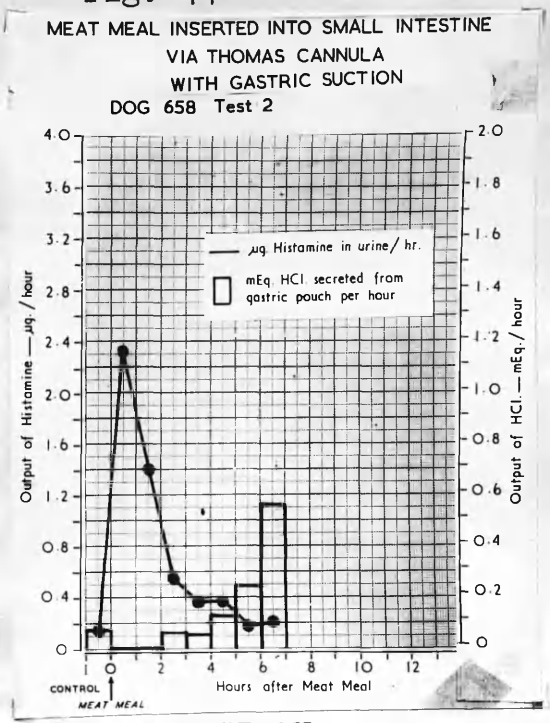


Fig. 44



MEAT INTO SMALL BOWEL WITHOUT GASTRIC SUCTION

Fig. 45

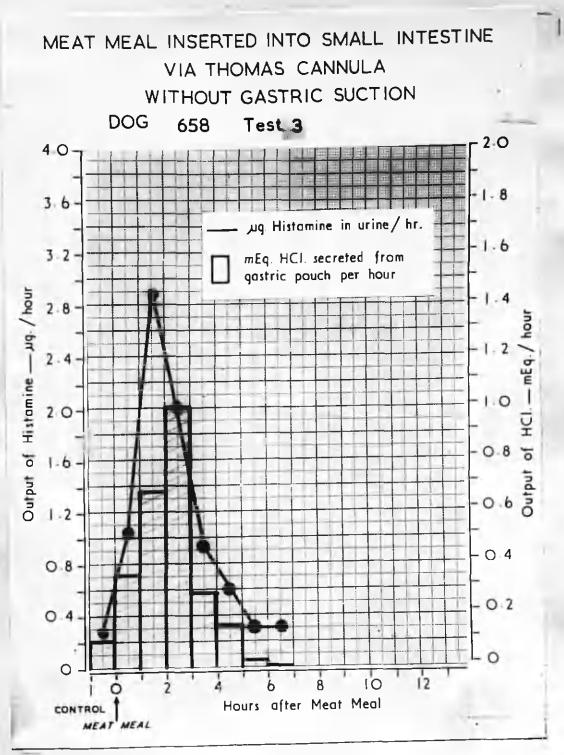
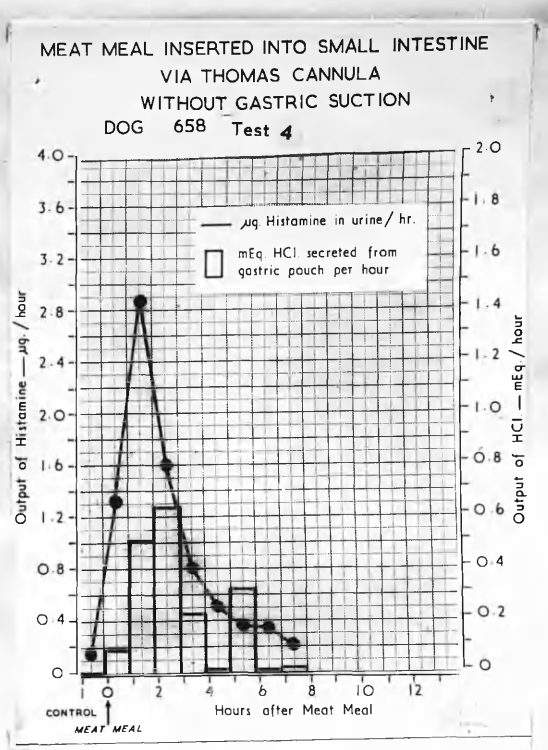


Fig. 46



Data from which graphs for dog 658 were constructed.

(Figs. 43 - 46).

Fig. 47.

EXP.	SPECIMEN	TOTAL URINE VOL ml	URINE VOL ON DECALSO COLUMN ml	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN. ml	HOURLY URINARY HISTAMINE µg	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq HCl per hr.
	CONTROL HOUR 1	125	50	10	0·4	0·03	0·17	2·2	0·0
	CONTROL HOUR 2	162	50	"	0·4	0·02	0·16	1·8	0·0
6	HOUR 1	120	50	"	0·29	0·2	1·6	0·5	0·0
5	HOUR 2	123	50	"	0·22	0·2	2·2	2·4	0·09
8	HOUR 3	64	50	"	0·19	0·12	1·08	10·7	0·94
TEST	HOUR 4	40	40	"	0·2	0·1	0·5	4·3	0·47
	HOUR 5	91	50	"	0·24	0·06	0·45	10·5	1·16
	HOUR 6	59	59	"	0·16	0·06	0·37	14·0	1·62
1	HOUR 7	56	56	"	0·12	0·04	0·33	16·0	1·39
	HOUR 8								
	HOUR 9			HISTAMINE STANDARD — 0·1 µg. / ml.					

EXP.	SPECIMEN	TOTAL URINE VOL ml	URINE VOL ON DECALSO COLUMN ml	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN. ml	HOURLY URINARY HISTAMINE µg	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq HCl per hr.
	CONTROL HOUR	146	50	10	0·4	0·02	0·146	0·6	0·07
	HOUR 1	89	50	"	0·06	0·08	2·32	0·2	0·0
6	HOUR 2	70	50	"	0·08	0·08	1·4	0·6	0·0
5	HOUR 3	121	50	"	0·4	0·07	0·434	1·3	0·06
8	HOUR 4	63	50	"	0·36	0·1	0·352	1·4	0·05
TEST	HOUR 5	65	50	"	0·36	0·1	0·36	2·3	0·12
	HOUR 6	33	33	"	0·3	0·05	0·17	4·0	0·24
	HOUR 7	48	48	"	0·3	0·06	0·2	5·9	0·56
2	HOUR 8								
	HOUR 9			HISTAMINE STANDARD — 0·1 µg. / ml.					
	HOUR 10								

EXP.	SPECIMEN	TOTAL URINE VOL. ml	URINE VOL ON DECALSO COLUMN ml	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN. ml	HOURLY URINARY HISTAMINE µg	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq HCl per hr.
	CONTROL HOUR	25	25	10	0·22	0·05	0·27	1·8	0·11
	HOUR 1	70	50	-	0·06	0·05	1·14	4·4	0·36
6	HOUR 2	120	50	-	0·05	0·05	2·88	5·8	0·68
5	HOUR 3	57	50	-	0·14	0·06	2·01	9·4	1·05
8	HOUR 4	62	50	-	0·08	0·06	0·93	3·0	0·28
	HOUR 5	38	38	-	0·1	0·06	0·6	2·0	0·16
TEST	HOUR 6	42	42	-	0·12	0·04	0·3	1·0	0·02
	HOUR 7	37	37	-	0·12	0·04	0·3	2·0	0·0
3	HOUR 8								
	HOUR 9			HISTAMINE STANDARD — 0·1 µg. / ml.					
	HOUR 10								

EXP.	SPECIMEN	TOTAL URINE VOL. ml	URINE VOL ON DECALSO COLUMN ml	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN. ml	HOURLY URINARY HISTAMINE µg	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq HCl per hr.
	CONTROL HOUR	40	40	10	0·4	0·06	0·15	0·9	0·0
	HOUR 1	50	50	-	0·06	0·08	1·33	1·7	0·08
6	HOUR 2	108	50	-	0·06	0·08	2·87	5·2	0·51
5	HOUR 3	100	50	-	0·1	0·08	1·6	5·9	0·63
8	HOUR 4	48	48	-	0·1	0·08	0·8	2·2	0·23
	HOUR 5	31	31	-	0·16	0·08	0·5	0·5	0·01
TEST	HOUR 6	43	43	"	0·22	0·08	0·36	3·3	0·3
	HOUR 7	65	50	"	0·30	0·08	0·34	1·3	0·02
	HOUR 8	50	50	"	0·30	0·06	0·2	2·5	0·02
4	HOUR 9								
	HOUR 10			HISTAMINE STANDARD — 0·1 µg. / ml.					

Dog 201

MEAT INTO SMALL BOWEL WITH GASTRIC SUCTION

Fig. 48

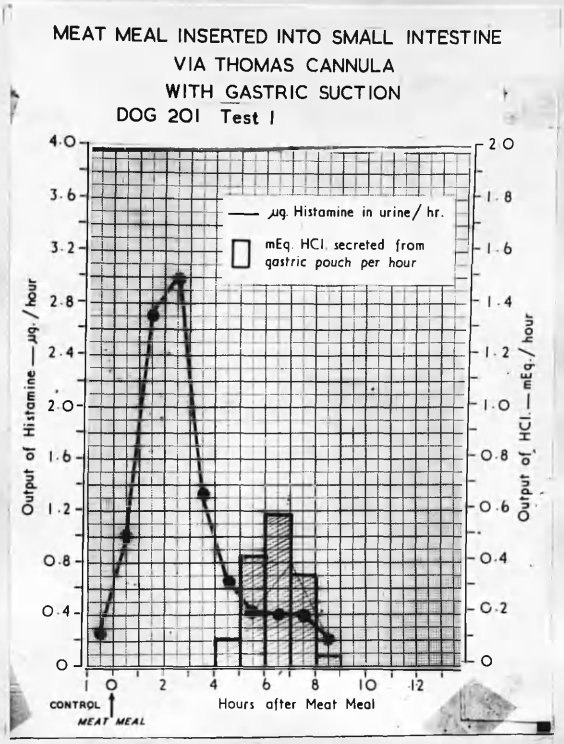
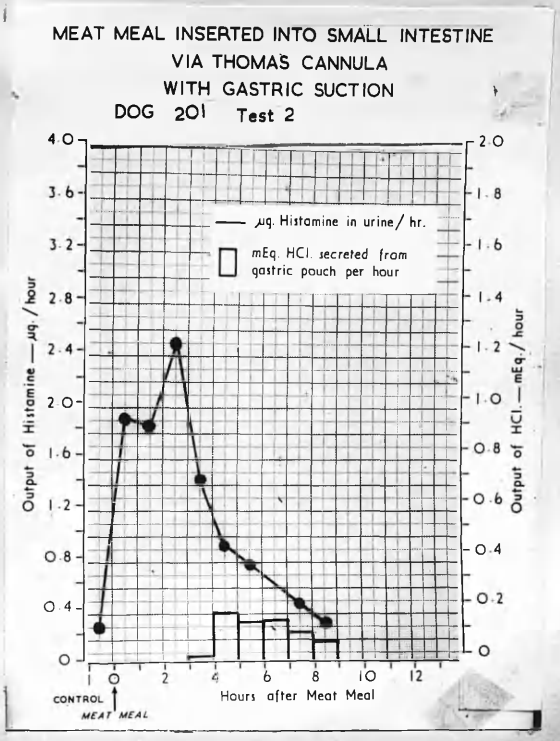


Fig. 49



EXP.	SPECIMEN	TOTAL URINE VOL. ml	URINE VOL ON DECALSO COLUMN ml	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL. UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN. ml	HOURLY URINARY HISTAMINE μg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
201 TEST 1.	CONTROL HOUR	70	50	10	0·3	0·06	0·28	1·8	0·0
	HOUR 1	69	50	"	0·08	0·06	1·03	1·5	0·0
	HOUR 2	230	50	"	0·1	0·06	2·72	1·0	0·0
	HOUR 3	198	50	"	0·08	0·06	3·0	1·8	0·0
	HOUR 4	102	50	"	0·09	0·06	1·32	2·8	0·0
	HOUR 5	72	50	"	0·13	0·06	0·66	2·9	0·11
	HOUR 6	50	50	"	0·14	0·06	0·42	5·2	0·43
	HOUR 7	30	30	"	0·12	0·08	0·4	6·5	0·59
	HOUR 8	30	30	"	0·12	0·08	0·4	5·0	0·35
	HOUR 9	50	50	"	0·4	0·08	0·2	1·4	0·04
	HOUR 10								

HISTAMINE STANDARD — 0·1 μg./ml.

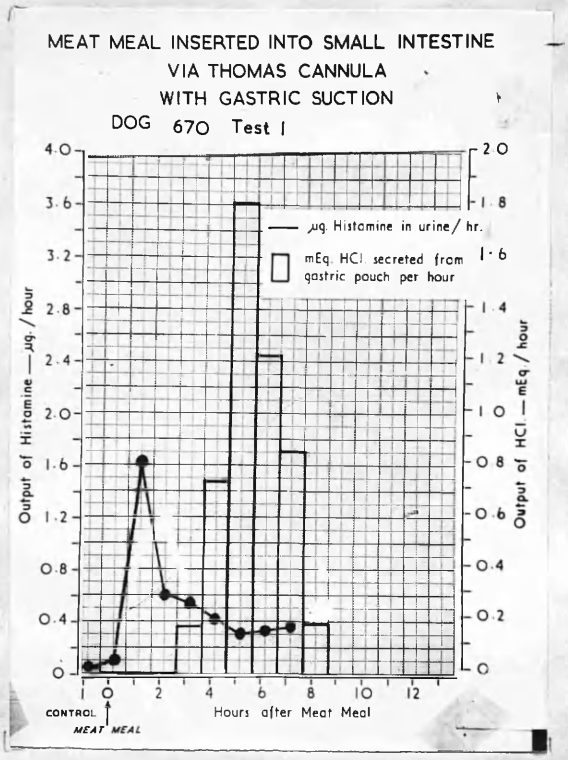
EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE μg.	VOL GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
670 TEST 1.	CONTROL HOUR	40	40	10	0·4	0·2	0·05	1·1	0·02
	HOUR 1	50	50	"	0·2	0·2	0·1	1·0	0·0
	HOUR 2	81	50	"	0·2	0·2	1·62	1·5	0·0
	HOUR 3	50	50	"	0·17	0·1	0·6	1·3	0·0
	HOUR 4	55	56	"	0·19	0·1	0·53	3·0	0·18
	HOUR 5	30	30	"	0·24	0·1	0·42	7·7	0·73
	HOUR 6	22	22	"	0·32	0·1	0·31	15·2	1·8
	HOUR 7	46	46	"	0·30	0·1	0·33	10·8	1·2
	HOUR 8	28	28	"	0·28	0·1	0·35	8·5	0·85
	HOUR 9	36	36	"	0·4	0·1	0·25	2·5	0·18
	HOUR 10								

HISTAMINE STANDARD — 0·1 μg./ml.

Dog 670

MEAT MEAL INTO SMALL BOWEL WITH GASTRIC SUCTION

Fig. 50



EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN ml.	HOURLY URINARY HISTAMINE μg.	VOL. GASTRIC JUICE SECRETED ml/hr.	mEq. HCl per hr.
201 Test 2	CONTROL HOUR	99	50	10	0·4	0·05	0·3	2·0	0·0
	HOUR 1	155	50	-	0·16	0·1	1·92	0·9	0·0
	HOUR 2	103	50	-	0·11	0·1	1·85	0·6	0·0
	HOUR 3	100	50	-	0·08	0·1	2·5	1·8	0·0
	HOUR 4	87	50	-	0·12	0·1	1·44	1·1	0·03
	HOUR 5	75	50	-	0·16	0·1	0·93	3·5	0·2
	HOUR 6	93	50	-	0·24	0·1	0·78	3·0	0·16
	HOUR 7	148	50	-	0·4	0·04	0·3	3·0	0·17
	HOUR 8	97	50	-	0·4	0·1	0·48	3·0	0·14
	HOUR 9	61	50	-	0·36	0·1	0·33	2·5	0·09
	HOUR 10								

HISTAMINE STANDARD — 0·1 μg. / ml.

COMMENTARY ON RESULTS.

When a meat meal was introduced into the upper jejunum of 4 dogs with Heidenhain pouches the changes in output of free histamine in the urine followed a uniform pattern in all tests. From low control values of 0.15 - 0.25 ug/hour, the output increased rapidly, peaks of around 2 ug/hour or more being reached in the second hour. While the output of histamine in the urine in the second hour was usually as great or greater than the peak reached following oral administration of the same amount of meat, this high output was not maintained over several hours as in the oral tests. Instead, in nearly all tests with intestinal feeding, the output of free histamine in the urine fell after the second or third hour and values approximating those obtained in the control hour were frequently obtained by the 4th hour.

Mean results obtained in two dogs are contrasted with the effect of an oral meal, meat in the isolated stomach, and intravenous infusions of histamine in Fig. 51. /

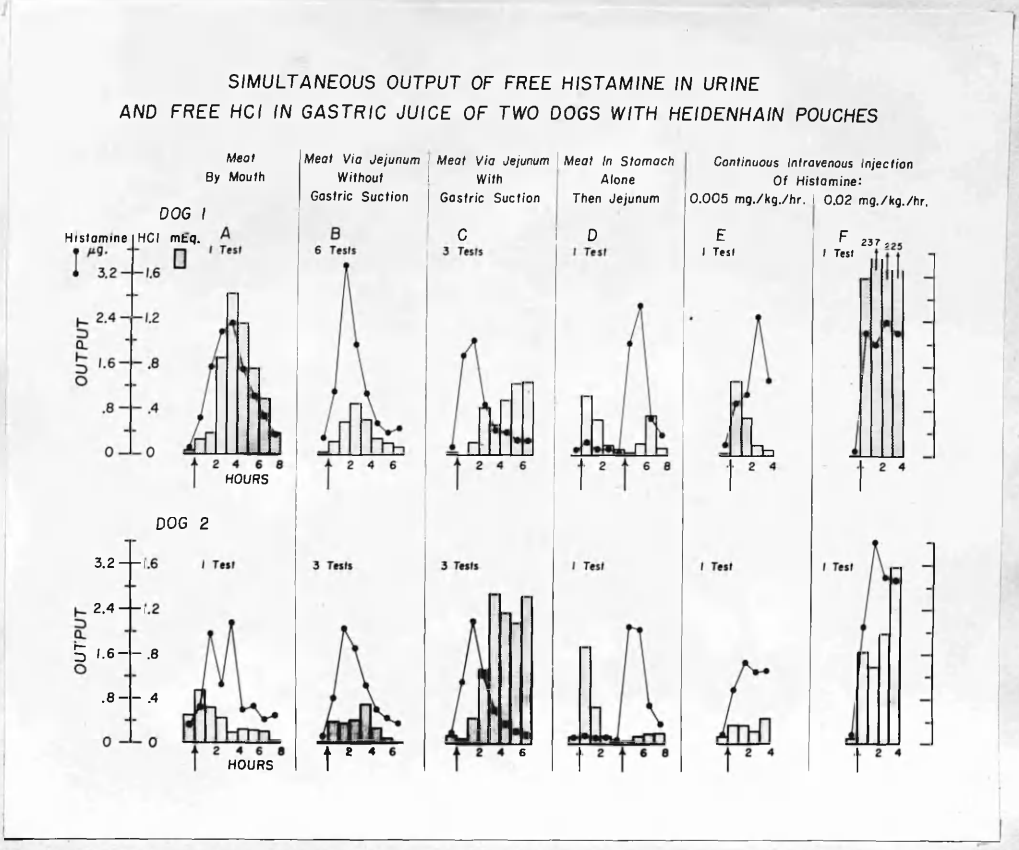


FIG. 51

The maximal acid response from the Heidenhain pouches of these dogs did not correspond to the maximal output of histamine in the urine, occurring as it did, usually between the 4th and 8th hours after the jejunal feed, when the urinary histamine had returned to a very low level. It will be noted that in tests without gastric suction more acid was usually secreted in the first two hours than was seen in similar tests with gastric suction, suggesting that some of this early acid secretion resulted from antral stimulation by the regurgitating meat. /

/meat.

The use of gastric suction reduced the acid output in the first two hours following the meat meal to low levels and, in some tests, no acid was secreted during this period when the urinary histamine reached its peak. This delayed acid response to intestinal feeding of 1 - 3 hours is in keeping with the observations of others (Chapter I). No statistical analysis is required to support the conclusion that the intestinal phase of acid secretion is not related to the elevation of urinary histamine occurring in these tests.

The question arises, why did the pouches not respond during the high peaks of free histamine in the urine which occurred in the second hour after the jejunal feed? If these elevated levels of free urinary histamine did in fact reflect a similar rise in free blood histamine, considerable acid secretion might have been expected in the second hour, yet almost none occurred. Would intravenous infusions of histamine, producing an overflow in the urine of the same magnitude as the peaks reached in the second hour, have stimulated the gastric pouches markedly? To clarify this point, each dog had a series of I.V. infusions of histamine at fixed rates and different dose levels to determine the acid response of the pouch when the output of free histamine in the urine was below and above that /

/that obtained in the second hour after the meat meal. Comparisons between the acid and urinary histamine response obtained by the meat feeds and intravenous infusions of histamine have already been illustrated in two of these dogs (Fig. 51). A similar comparison for a third dog is illustrated in Fig. 52.

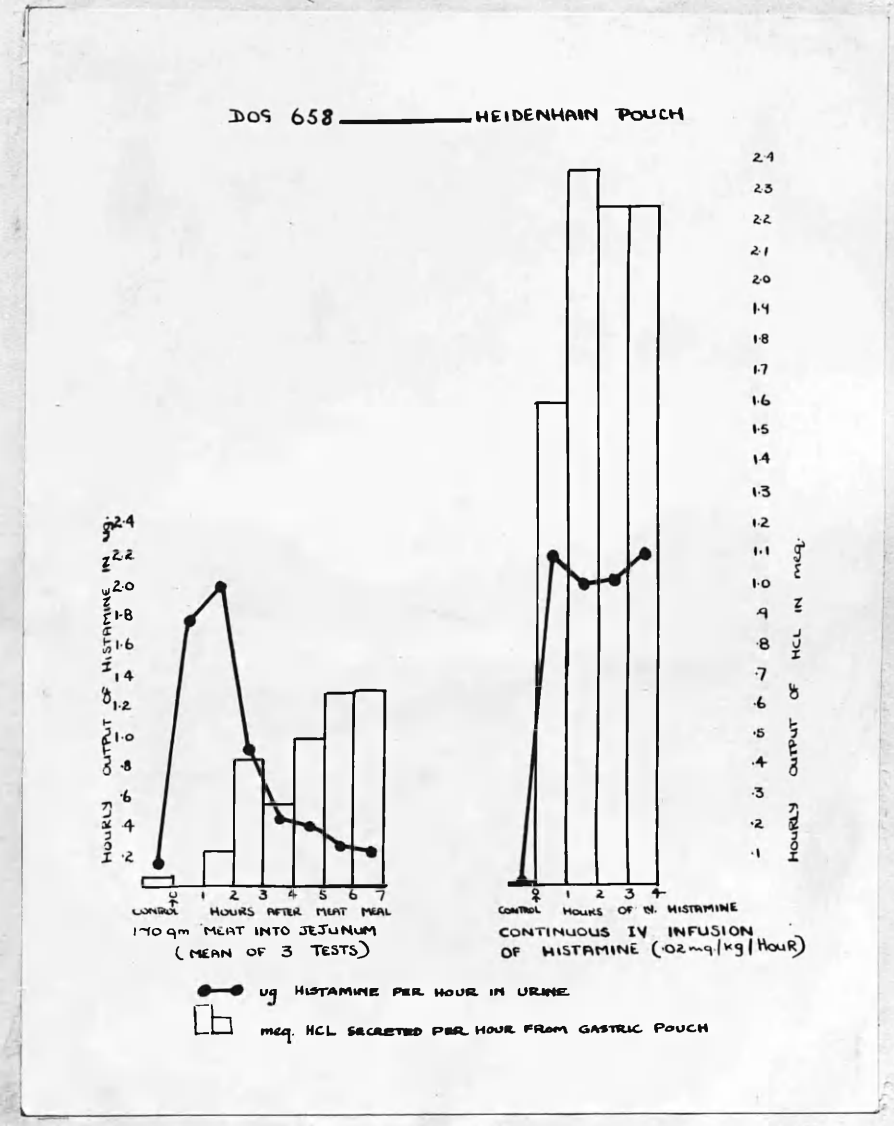


FIG. 52

One must conclude that if the peak of free histamine excretion in the second hour following the intestinal meal of meat reflected a similar rise in free blood histamine, as was the case with the I.V. histamine tests, a significant augmentation in acid secretion from the pouches should have occurred. Is it possible that such an elevation in free blood histamine did occur but that in the early hours after the feed, some inhibitor mechanism was at work preventing the pouch from responding to histamine. To test this hypothesis, one dog was given a subcutaneous dose of histamine and the acid secretion from the pouch measured. The usual homogenised meat meal was then placed in the small bowel and the dose of subcutaneous histamine repeated. In the 3 hour period following the intestinal meat meal a second similar acid response was obtained from the administration of subcutaneous histamine. No evidence was obtained that in the 3 hours following the entrance of meat into the small bowel any inhibition of the response of the pouch to histamine occurred. (Fig. 53).

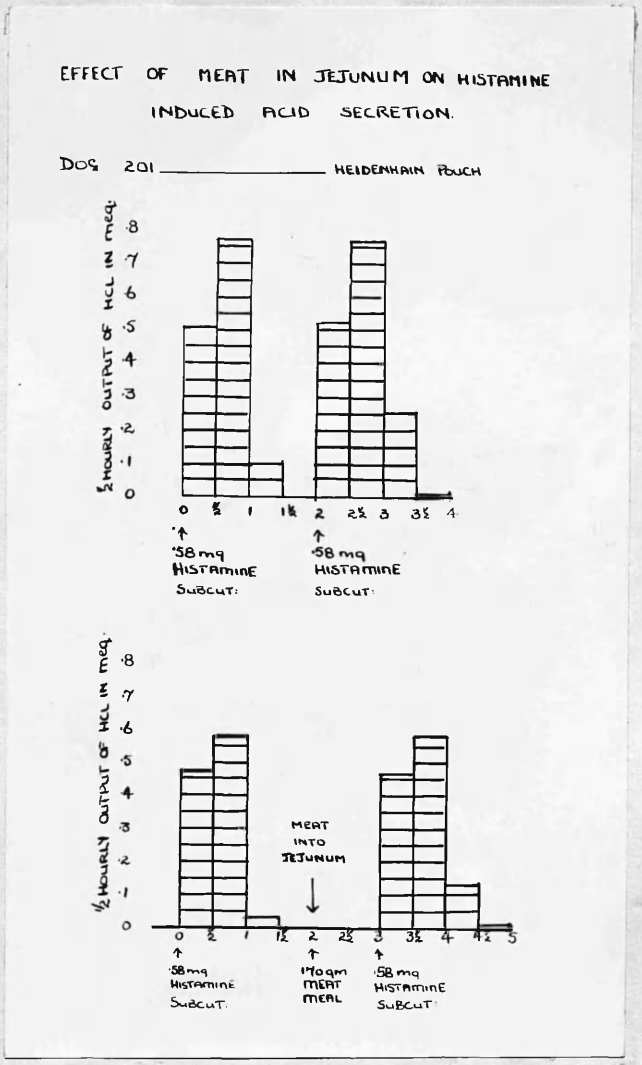


FIG. 53.

Why then did the pouch not secrete histamine during the second hour peak in the urine? When parenteral histamine produced a similar urinary output, the pouches strongly secreted. There was no evidence that the meat in the small bowel inhibited the pouches' acid response /

/response to histamine.

Is it possible that meat in the small bowel produces a rise in inactive, loosely bound histamine in the blood, which is removed by the kidney and excreted in the free form?

This raises the question of the form in which histamine circulates in the blood.

IS HISTAMINE FREE IN THE BLOOD?

What evidence is there that histamine does exist in a free form in the plasma under physiological conditions? Kaiser (1939) was of the opinion that histamine was bound to protein and inactive. This theory that histamine combines loosely and reversibly with proteins has been tested by experiments involving dialysis or ultra-filtration and the results do not support it (Emmelin, 1945: Boon and Vane, 1952: Kaplan and Davis, 1953).

Rocha e Silva (1946) has suggested that histamine may be united to tissue proteins by peptide linkage to a carboxyl group, since histamine is liberated from tissues by pure crystalline pepsin. The trypsin might produce this result by acting on the peptide linkage but again it may do so by destroying the cell structure. Apparently Lindahl (1954) obtained some evidence that histamine might form an ether soluble compound with lecithin. /

/lecithin.

Obrink (1948) was of the opinion that histamine probably occurred in an inactive form in the plasma. In his dog experiments he found high levels of whole blood histamine with no acid secretion. There was no evidence that the gastric glands were in some way adapted to the actual histamine concentration and started secreting only when the level of histamine in the blood was increased. However, most of the blood histamine is intracellular, in close relation to the mitochondria and anatomical factors may have prevented it from acting on the gastric glands.

One might summarise this information by saying that many observers have suggested that histamine occurs in a loosely bound inactive form in the blood under physiological conditions but that no strong evidence exists to support this view (Gaddum, 1956). The only bound histamine definitely known to occur is acetyl histamine (Chapter II) and this compound is quite stable, requiring prolonged boiling with acid to liberate the free form. On the other hand it seems very difficult to disprove the cross-circulation experiments of Emmelin (1945), suggesting that histamine occurred in a free form in the blood of guinea pigs as measured by the bronchial constriction reaction. The experiments in this chapter might be taken as support /

/support for the concept that increases in blood histamine occur in an inactive form under physiological conditions. Intravenous infusions of histamine on the other hand appear to produce a rise in free blood histamine as measured by the acid response of the pouch and overflow in the urine.

Another explanation of these results must be considered. Did the meat in the small bowel produce an increased excretion of free histamine in the urine without any change in the blood level taking place? No information is available to exclude completely this possibility.

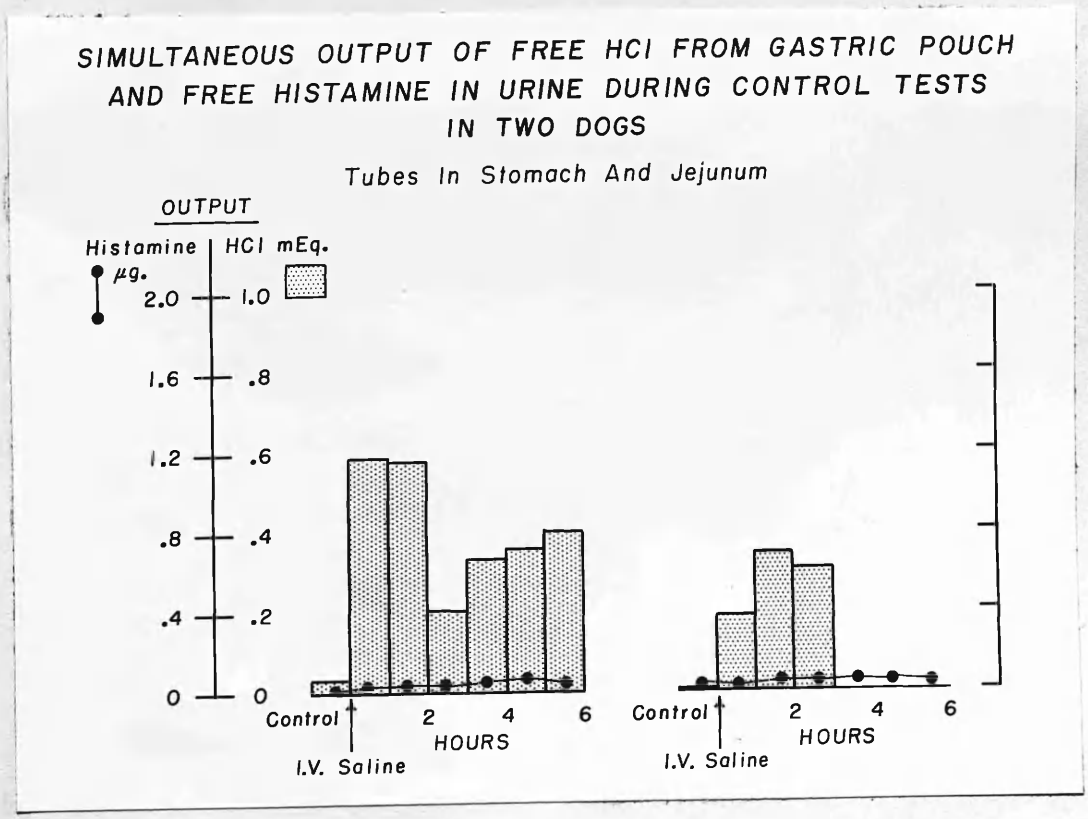
How certain can one be that the substance which appears in the urine is histamine? It is not possible, short of complete chemical identification, to prove conclusively that the gut contracting substance present in the urine is histamine. It can be said that it is indistinguishable from histamine over a wide range of tests, both quantitatively and qualitatively.

CONTROL STUDIES WITH SHAM INTESTINAL MEAL.

In these studies of intestinal secretion, the homogenised meat was introduced into the jejunum along a tube passed through the Thomas cannula. The manoeuvre involved considerable manipulation and mechanical stimulation to the gut. It was necessary to exclude the possibility that the manipulations alone augmented the urinary histamine. Two dogs/

/dogs were therefore prepared in the usual way for a typical test. The bladders were catheterised and continuous infusions of hypotonic saline commenced. After a control hour a tube was passed into the Thomas cannula, but on this occasion no meat was installed. Gastric suction was applied for three hours as in previous tests. The results obtained are illustrated in Fig. 54. A study of this illustration shows clearly that the manipulations and intravenous infusions did not increase the free histamine in the urine. On the other hand, these disturbances did appear to stimulate some acid secretion. This must be taken into account when studying the magnitude of the acid response obtained in these experiments.

Fig. 54



What then causes the excretion of free histamine in the urine, if histamine it is, after meat enters the small bowel. Apparently this effect is peculiar to meat. When in each dog, a bread and milk meal of identical calorific value and volume to the meat meal, was introduced into the jejunum through the Thomas cannula, no rise in free histamine in the urine occurred. (Fig. 55 see over).

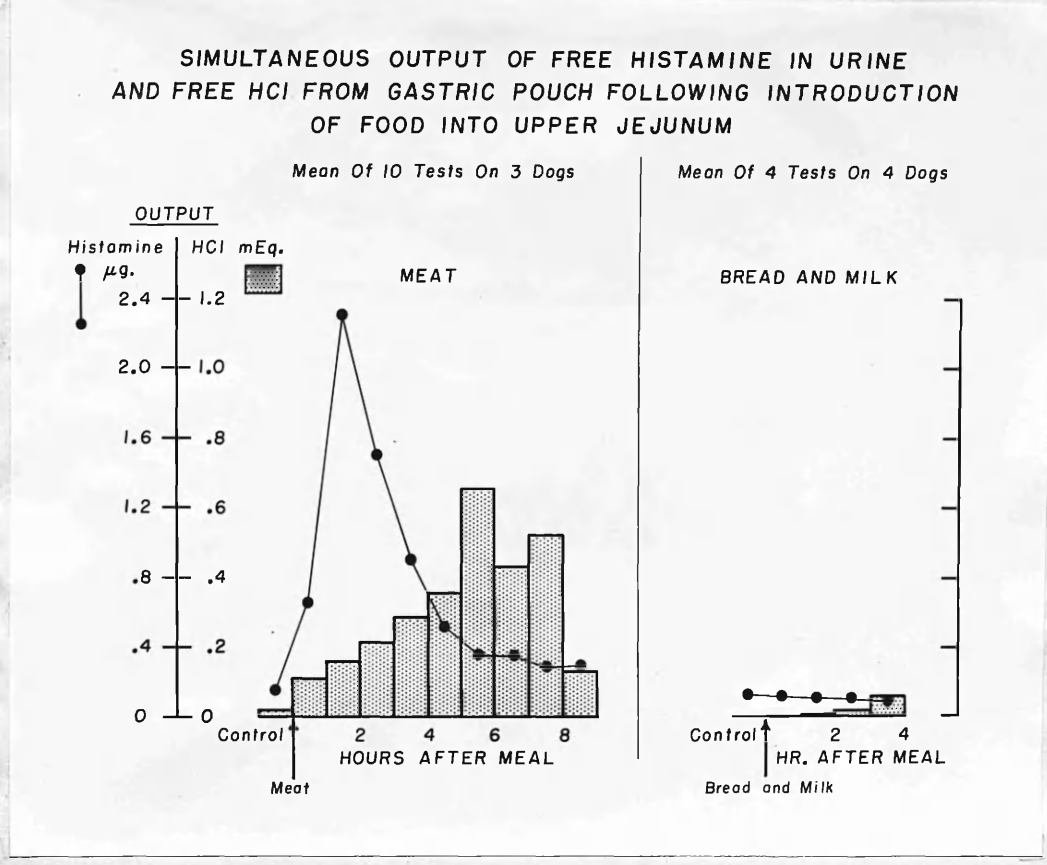


FIG. 55

In the chapter which follows an effort has been made to find a substance in the meat meal which might be a possible source of this urinary histamine.

SUMMARY OF CHAPTER.

(1) The free histamine in the urine and acid output from Heidenhain pouches has been measured before and after the introduction of a meat meal into the upper jejunum of dogs. /

/dogs.

(2) After the introduction of a meat meal into the jejunum, there is a rapid rise in the free urinary histamine followed by a rapid fall, control levels being reached by the 4th hour in most experiments.

The acid response is almost absent in the first 3 hours and is maximal between the 4th and 6th hours when urinary histamine output has again reached control levels.

(3) There would appear to be no relationship between the output of free histamine in the urine after a meat meal and the intestinal phase of acid secretion.

(4) The experiments described produce evidence that might support the concept that histamine may occur in the blood stream in an inactive bound form.

(5) The appearance of free histamine in the urine which follows when meat enters the small bowel appears to be peculiar to meat. A bread and milk meal of equal calorific value, produces no change in the urinary histamine.

CHAPTER IX.

L-HISTIDINE AS A POSSIBLE SOURCE OF THE INCREASE OF FREE HISTAMINE IN THE URINE AFTER A MEAT MEAL.

In the two previous chapters it was seen that the rise in free histamine in the urine which followed a meal of meat, only occurred when the meat entered the small bowel. This ability of a meat meal to augment the urinary histamine was not shared by a bread and milk meal given under identical conditions. It produced no such rise. What substances in the meat could have effected this change? Meat itself contains small amounts of histamine. Skeletal muscle of horse origin may have 20-40 ug/gm. and meat derived from cattle 1-10 ug/gm (Feldberg, 1956). Thus a meat meal of 150 gms. might contain 1.5 - 6 mg. of histamine at most and the content may well be as low as 0.15 mg - 3 mg. As will be seen in the next chapter, this is unlikely, in itself, to account for all the free histamine in the urine after a meat meal.

Meat contains considerable quantities of L-histidine (2-4%, Tabor, 1954) and it is well known that bacteria form histamine from histidine by decarborylation (Ackerman, 1910, 1911: Berthelot and Bertrans, 1912: Mellanby and Twort, 1912: Kendall and Gebauer, 1930: Matsouda, 1933: Gale, 1940: Epps, 1945). Another /

/Another potential source of histamine in meat is carnosine, a peptide of L-histidine and β alanine. On acid hydrolysis it would yield L-histidine. However its concentration in muscle is very small (100 - 1000 micrograms/gm. wet tissue - Tabor, 1954) and this mechanism would only add a relatively insignificant amount of histidine to that already available for decarboxylation. The high cost of carnosine also prohibits its extensive use in experimental work.

It was therefore considered possible that decarboxylation of L-histidine in the meat might produce histamine in the alimentary tract which was absorbed and excreted in the urine. Such a concept would account for the rise in the urinary histamine occurring only after the meat had entered the small bowel, since, for decarboxylation, an alkaline media and the presence of bacteria would be required.

If such was the explanation, then reduction of the intestinal flora should reduce the output of free histamine in the urine following a meal of meat. Wilson (1954), has demonstrated that a number of drugs differing widely from each other in chemical structure (chloramphenicol, aureomycin, sulphasuccidine) but having in common the capacity to reduce the intestinal flora, all reduce the excretion of free histamine in the rat.

In this chapter a series of experiments are reported with two objectives. /

/objectives.

(1) To compare the output of free histamine in the urine after a meat meal with that obtained by giving its estimated histidine content in pure form.

(2) To study the effect of sterilising the gut on the urinary histamine response to meat and L-histidine.

MATERIAL AND METHODS.

Most of these studies were carried out in adult patients convalescing from herniorrhaphy or uncomplicated appendicectomy.

Experiments followed a 16-hour fast and consisted of hourly collections of urine before and after administration of the meal. The meat meal consisted of 150 gms. of cooked finely minced meat. The histidine meal consisted of 4.5 gms. L-histidine suspended in milk. Since the histidine content of meat has been taken as 2-4% (Tabor, 1954) the amount chosen was 3% of the 150 gm. meat meal. In one test 2 gms. of Histamine was given by mouth. Urinary output was maintained by small amounts of water by mouth, and collections were made by normal evacuation of the bladder.

RESULTS. The tests carried out in the four patients are recorded in the pages which follow. Where multiple graphs are placed on a single page, the tables from which the graphs are drawn follow immediately after.

Patient 1

Meat meal by mouth

Fig. 56

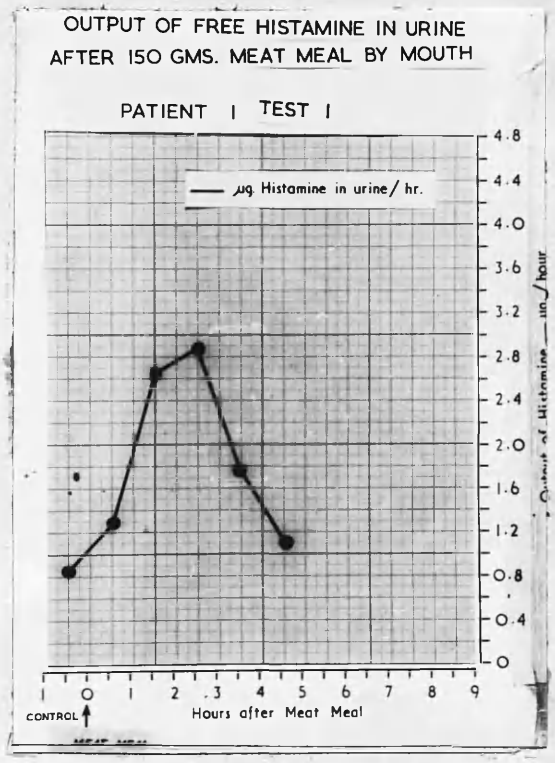
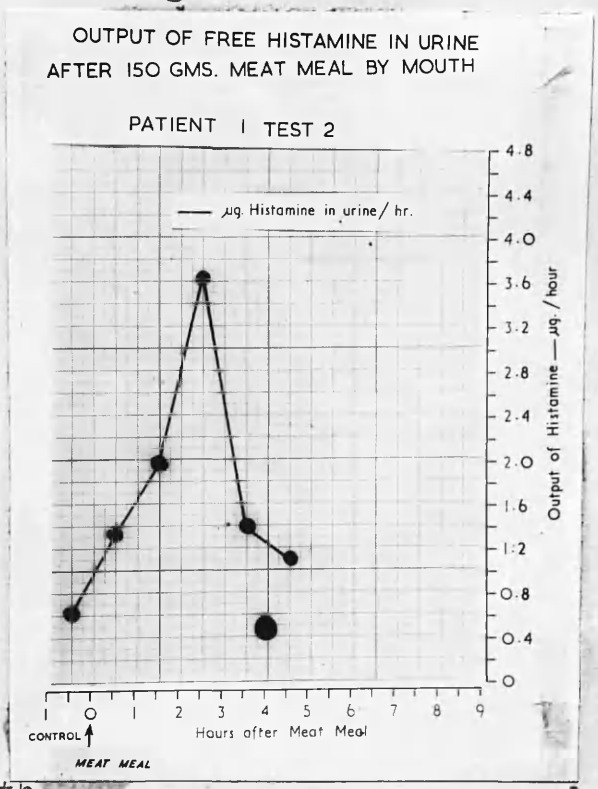


Fig. 57



L-Histidine by mouth

Fig. 58

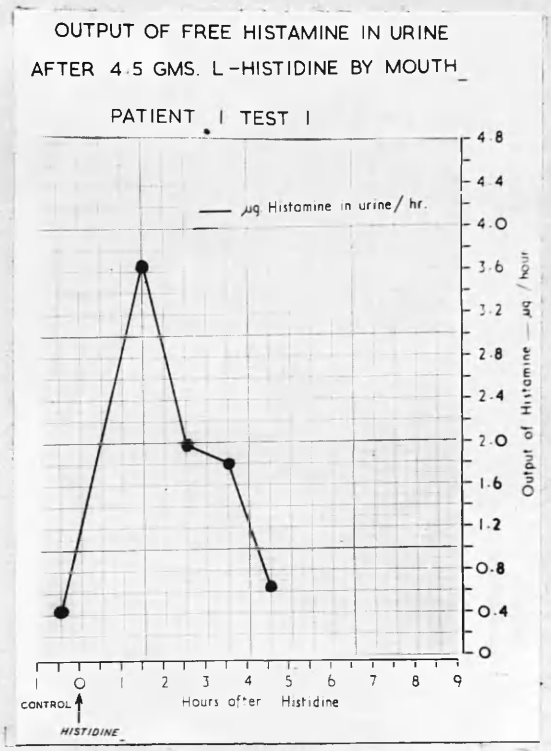
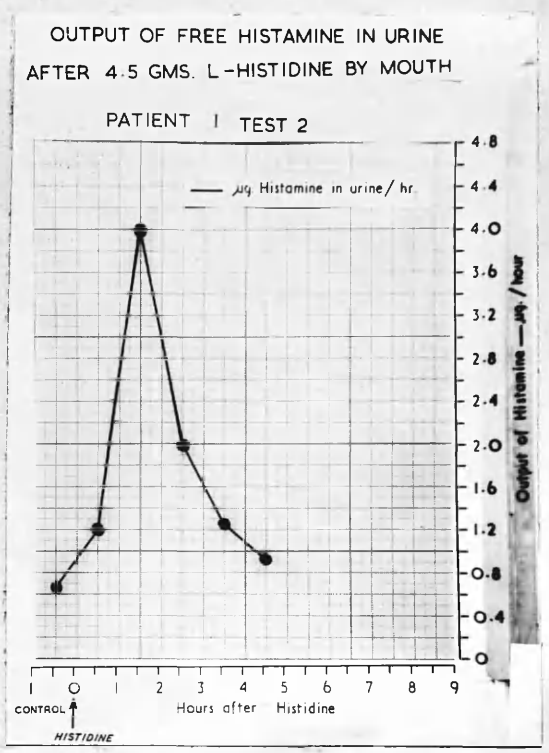


Fig. 59



Patient 1

Meat meal by mouth

OUTPUT OF FREE HISTAMINE IN URINE AFTER 150 GMS. MEAT MEAL BY MOUTH

PATIENT 1 TEST 1

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL GASTRIC JUICE SECRETED ml./hr.	mEq HCl per hr.
CONTROL HOUR	170	50	10	0·4	0·1	0·85		
HOUR	103	50	11	0·6	0·35	1·276		
HOUR	150	50	11	0·5	0·4	2·64		
HOUR	144	50	10	0·4	0·4	2·88		
HOUR	101	50	10	0·5	0·4	1·76		
HOUR	76	50	10	0·4	0·3	1·12		
HOUR								
HOUR								
HOUR								

HISTAMINE STANDARD — 0·1 µg. / ml.

OUTPUT OF FREE HISTAMINE IN URINE AFTER 150 GMS. MEAT MEAL BY MOUTH

PATIENT 1 TEST 2

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL GASTRIC JUICE SECRETED ml./hr.	mEq HCl per hr.
CONTROL HOUR	125	50	10	0·4	0·1	0·625		
HOUR 1	134	50	10	0·4	0·2	1·34		
HOUR 2	300	50	10	0·6	0·2	1·98		
HOUR 3	146	50	10	0·4	0·5	3·65		
HOUR 4	142	50	11	0·5	0·2	1·17		
HOUR 5	109	50	10	0·4	0·2	1·09		
HOUR 6								
HOUR 7								
HOUR 8								

HISTAMINE STANDARD — 0·1 µg. / ml.

L-Histidine by mouth

OUTPUT OF FREE HISTAMINE IN URINE AFTER 4·5 GMS. L-HISTIDINE BY MOUTH

PATIENT 1 TEST 1

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	65	50	10	0·6	0·2	0·429
HOUR 1	439	50	10	SPECIMEN LOST		
HOUR 2	165	50	10	0·35	0·4	3·76
HOUR 3	150	50	10	0·6	0·4	1·98
HOUR 4	90	50	10	0·4	0·4	1·8
HOUR 5	42	42	10	0·6	0·4	0·66
HOUR 6						

HISTAMINE STANDARD — 0·1 µg. / ml.

OUTPUT OF FREE HISTAMINE IN URINE AFTER 4·5 GMS. L-HISTIDINE BY MOUTH

PATIENT 1 TEST 2

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	100	50	10	0·6	0·2	0·66
HOUR 1	240	50	10	0·6	0·15	1·2
HOUR 2	100	50	10	0·2	0·4	4·0
HOUR 3	125	50	10	0·5	0·4	2·0
HOUR 4	255	50	10	0·4	0·1	1·25
HOUR 5	85	50	10	0·55	0·3	0·92
HOUR 6						

HISTAMINE STANDARD — 0·1 µg. / ml.

Patient 1

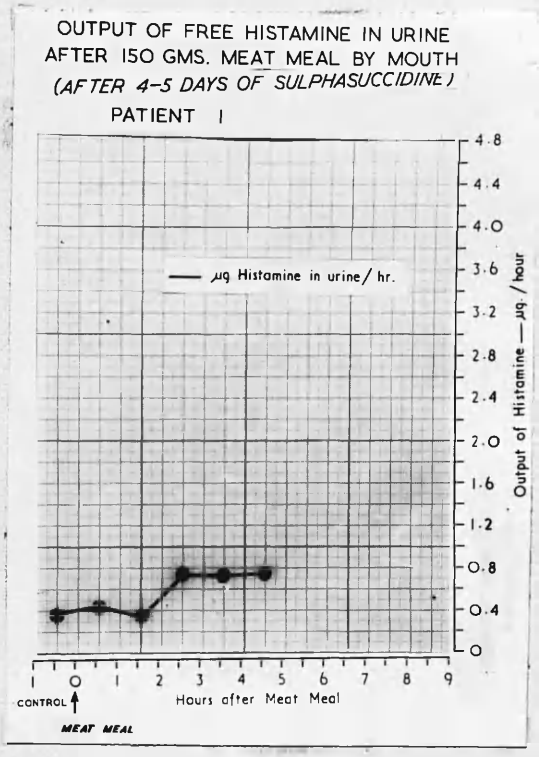
Meat meal by mouth after sulphasuccidine

Fig. 60

OUTPUT OF FREE HISTAMINE IN URINE
AFTER 150 GMS. MEAT MEAL BY MOUTH
(AFTER 4-5 DAYS OF SULPHASUCCIDINE)
PATIENT 1

SPECIMEN	TOTAL URINE VOL. ml	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL. UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN. ml	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	30	30	10	0·4	0·15	0·37
HOUR 1	48	48	10	0·35	0·15	0·43
HOUR 2	76	50	10	0·5	0·1	0·34
HOUR 3	100	50	10	0·4	0·15	0·74
HOUR 4	226	50	10	0·6	0·1	0·72
HOUR 5	210	50	10	0·55	0·1	0·75
HOUR 6						

HISTAMINE STANDARD — 0·1 µg. / ml.



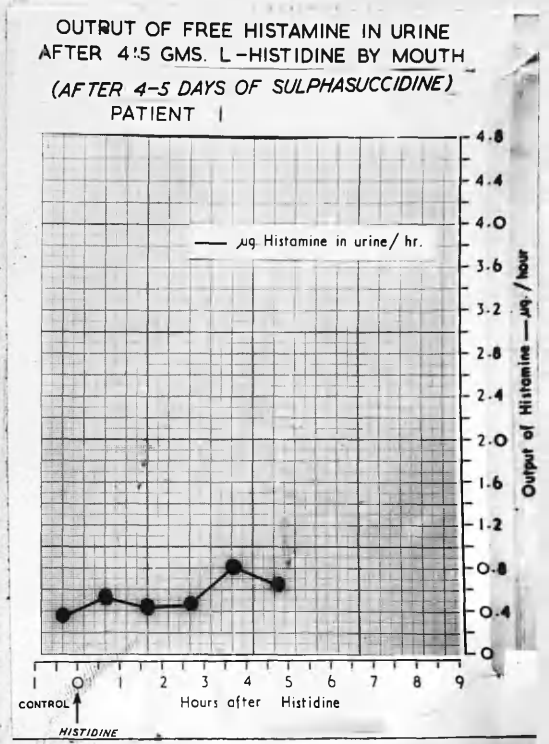
L-Histidine by mouth after sulphasuccidine

Fig. 61

OUTPUT OF FREE HISTAMINE IN URINE
AFTER 4·5 GMS. L-HISTIDINE BY MOUTH
(AFTER 4-5 DAYS OF SULPHASUCCIDINE)
PATIENT 1

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	166	50	10	0·6	0·1	0·34
HOUR 1	107	50	10	0·4	0·1	0·53
HOUR 2	88	50	10	0·4	0·1	0·44
HOUR 3	280	50	10	0·6	0·05	0·46
HOUR 4	250	50	10	0·6	0·1	0·80
HOUR 5	165	50	10	0·5	0·1	0·66
HOUR 6						

HISTAMINE STANDARD — 0·1 µg. / ml.



Patient 2

Meat meal by mouth

Fig. 62

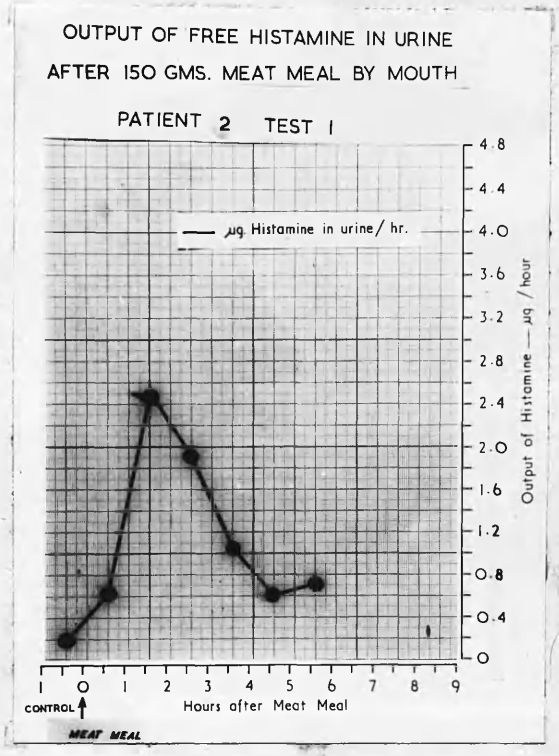
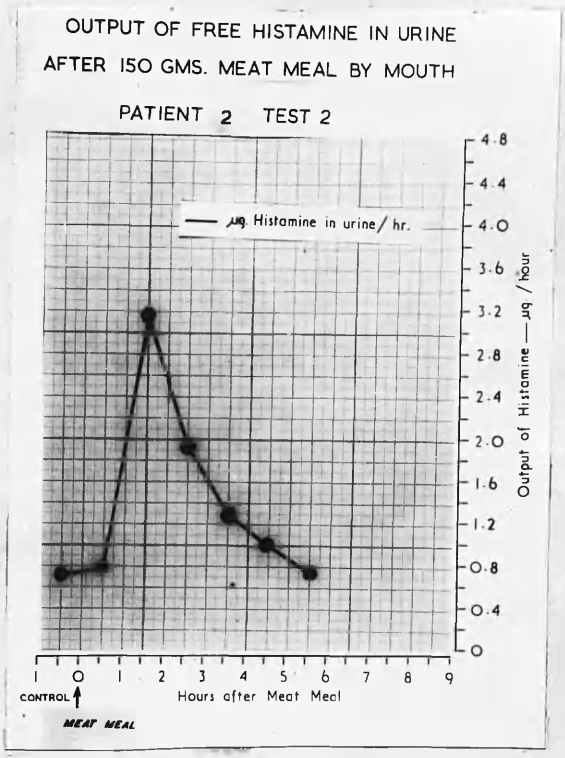


Fig. 63



L-Histidine by mouth

Fig. 64

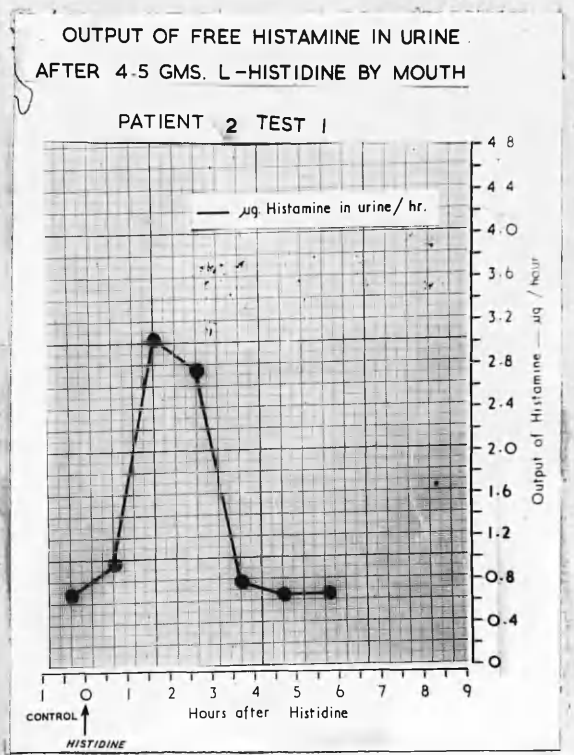
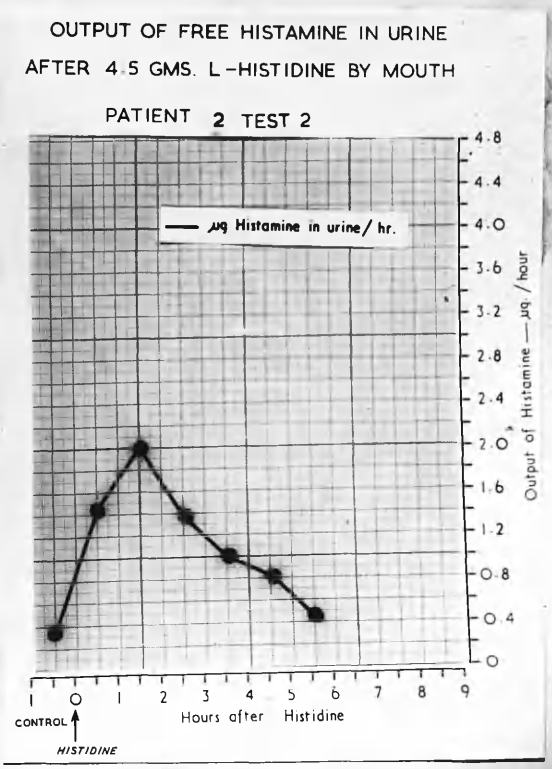


Fig. 65



Patient 2

Meat meal by mouth

OUTPUT OF FREE HISTAMINE IN URINE AFTER 150 GMS. MEAT MEAL BY MOUTH

PATIENT 2 TEST 1

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	34	34	10	0·5	0·1	0·20
HOUR 1	152	50	14	0·5	0·1	0·64
HOUR 2	250	50	10	0·4	0·2	2·5
HOUR 3	160	50	12·5	0·33	0·2	1·92
HOUR 4	70	50	12·5	0·33	0·25	1·05
HOUR 5	42	50	12·5	0·5	0·3	0·6
HOUR 6	54	50	12·0	0·5	0·3	0·72

HISTAMINE STANDARD — 0·1 µg. / ml.

OUTPUT OF FREE HISTAMINE IN URINE AFTER 150 GMS. MEAT MEAL BY MOUTH

PATIENT 2 TEST 2

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	90	50	12	0·6	0·2	0·72
HOUR 1	70	50	13	0·8	0·45	0·78
HOUR 2	90	50	12	0·3	0·5	3·16
HOUR 3	100	50	12	0·4	0·3	1·92
HOUR 4	64	50	13	0·4	0·3	1·28
HOUR 5	34	34	12·5	0·3	0·3	1·0
HOUR 6	76	50	12·5	0·6	0·3	0·76

HISTAMINE STANDARD — 0·1 µg. / ml.

L-Histidine by mouth

OUTPUT OF FREE HISTAMINE IN URINE AFTER 4·5 GMS. L-HISTIDINE BY MOUTH

PATIENT 2 TEST 1

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	48	48	10	0·3	0·2	0·67
HOUR 1	134	50	10	0·7	0·25	0·96
HOUR 2	152	50	10	0·3	0·3	3·04
HOUR 3	110	50	10	0·4	0·5	2·75
HOUR 4	76	50	10	0·6	0·3	0·76
HOUR 5	90	50	10	0·7	0·25	0·65
HOUR 6	76	50	10	0·7	0·3	0·65

HISTAMINE STANDARD — 0·1 µg. / ml.

OUTPUT OF FREE HISTAMINE IN URINE AFTER 4·5 GMS. L-HISTIDINE BY MOUTH

PATIENT 2 TEST 2

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	54	54	10	0·8	0·25	0·34
HOUR 1	180	50	10	0·5	0·2	1·44
HOUR 2	50	50	10	0·2	0·4	2·0
HOUR 3	86	50	10	0·5	0·4	1·37
HOUR 4	27	50	10	0·3	0·3	1·0
HOUR 5	33	33	10	0·37	0·3	0·81
HOUR 6	18	18	·10	0·7	0·3	0·43

HISTAMINE STANDARD — 0·1 µg. / ml.

Patient 2

Meat meal by mouth after sulphasuccidine

Fig. 66

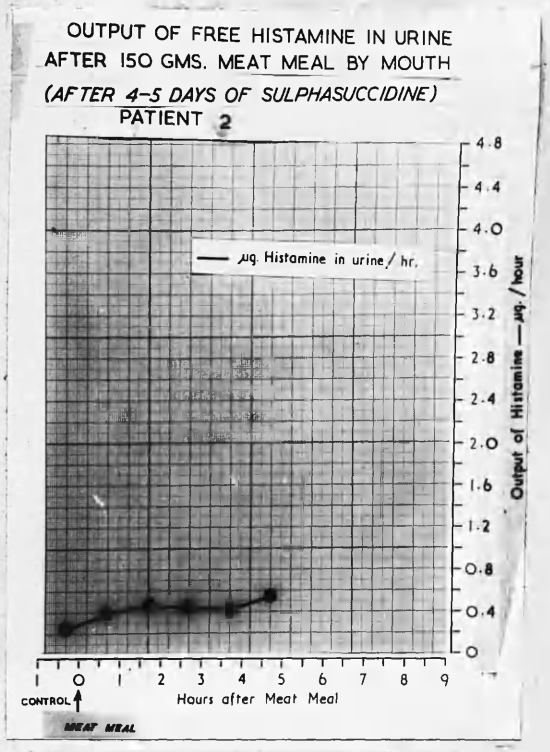
OUTPUT OF FREE HISTAMINE IN URINE AFTER 150 GMS. MEAT MEAL BY MOUTH

(AFTER 4-5 DAYS OF SULPHASUCCIDINE)

PATIENT 2

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	42	42	10	1	0·25	0·25
HOUR 1	64	50	10	1	0·3	0·384
HOUR 2	78	50	10	1	0·3	0·468
HOUR 3	55	50	10	1	0·4	0·44
HOUR 4	112	50	10	1	0·2	0·416
HOUR 5	90	50	10	1	0·3	0·54
HOUR 6						

HISTAMINE STANDARD — 0·1 µg. / ml.



L-Histidine by mouth after sulphasuccidine

Fig. 67

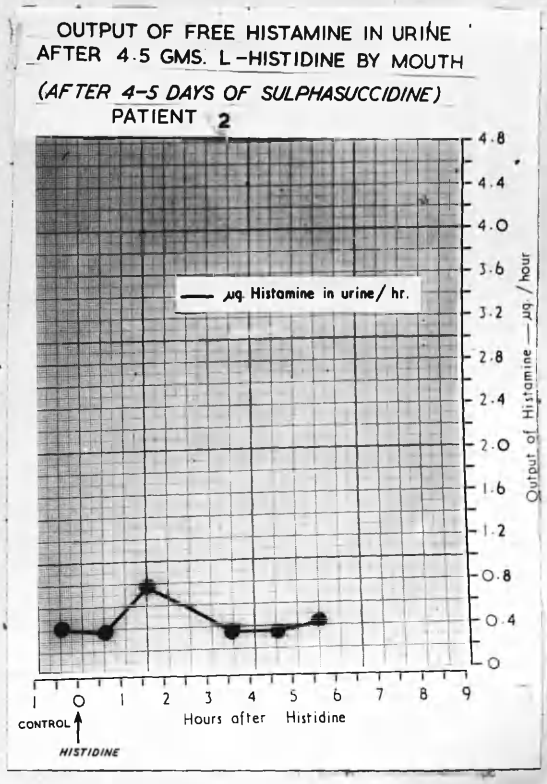
OUTPUT OF FREE HISTAMINE IN URINE AFTER 4·5 GMS. L-HISTIDINE BY MOUTH

(AFTER 4-5 DAYS OF SULPHASUCCIDINE)

PATIENT 2

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	48	48	10	0·8	0·3	0·4
HOUR 1	64	50	"	1·0	0·3	0·38
HOUR 2	192	50	"	1·0	0·2	0·77
HOUR 3	82	50	"	1·0	0·35	0·57
HOUR 4	50	50	"	1·0	0·35	0·35
HOUR 5	42	42	"	1·0	0·35	0·35
HOUR 6	72	50	"	1·0	0·3	0·43

HISTAMINE STANDARD — 0·1 µg. / ml.



Patient 3.

Meat meal by mouth.
Fig.68

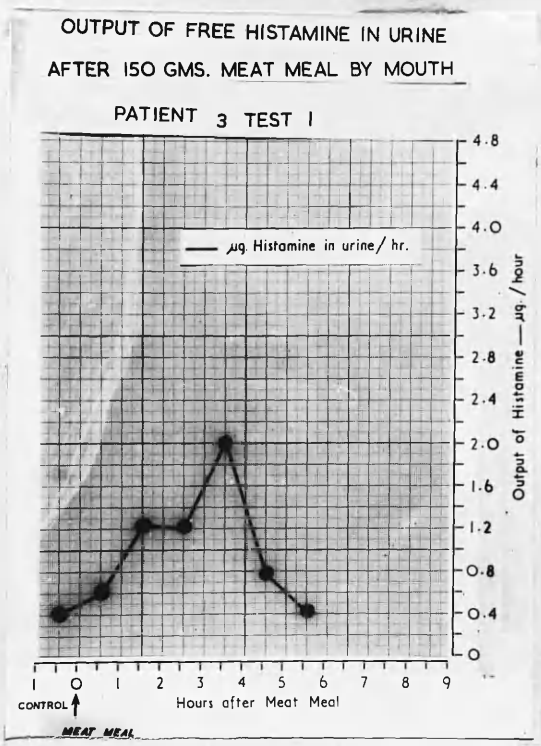
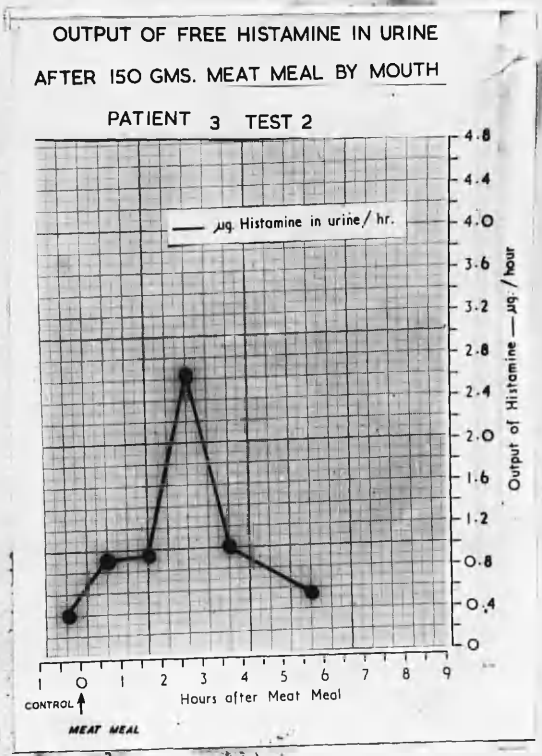
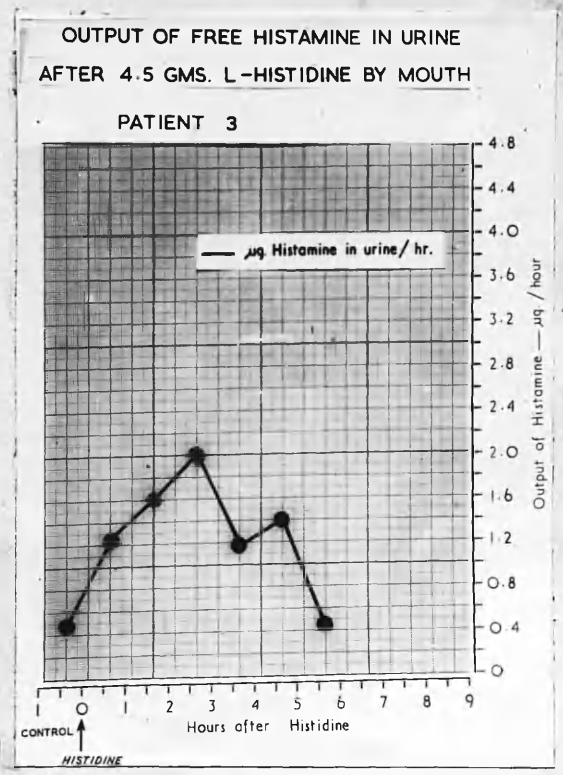


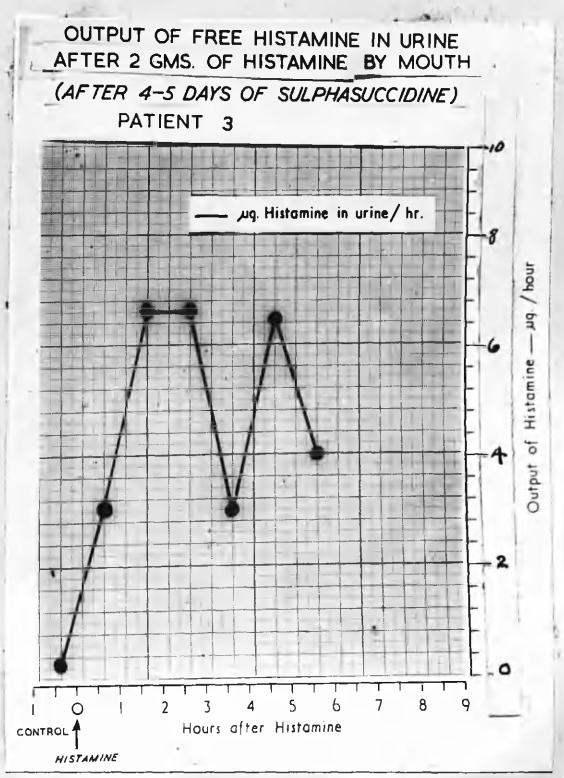
Fig.69



L-Histidine by mouth.
Fig.70



Histamine by mouth.
Fig.71



Patient 3.

Meat meal by mouth.

OUTPUT OF FREE HISTAMINE IN URINE AFTER 150 GMS. MEAT MEAL BY MOUTH

PATIENT 3 TEST 1

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	38	38	10	1·0	0·4	0·4
HOUR 1	36	36	10	1·0	0·6	0·6
HOUR 2	32	32	10	0·3	0·4	1·22
HOUR 3	100	50	10	0·33	0·2	1·2
HOUR 4	49	49	10	0·10	0·2	2·0
HOUR 5	56	50	10	0·5	0·35	0·78
HOUR 6	120	50	10	0·6	0·1	0·41

HISTAMINE STANDARD — 0·1 µg. / ml.

OUTPUT OF FREE HISTAMINE IN URINE AFTER 150 GMS. MEAT MEAL BY MOUTH

PATIENT 3 TEST 2

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	44	44	10	0·6	0·2	0·4
HOUR 1	70	50	10	0·6	0·4	0·92
HOUR 2	94	50	10	0·4	0·2	0·94
HOUR 3	88	50	10	0·5	0·6	2·64
HOUR 4	60	50	10	0·6	0·5	0·99
HOUR 5	110		SAMPLE	LOST		
HOUR 6	108	50	10	0·4	0·1	0·54

HISTAMINE STANDARD — 0·1 µg. / ml.

L-Histidine by mouth.

OUTPUT OF FREE HISTAMINE IN URINE AFTER 4·5 GMS. L-HISTIDINE BY MOUTH

PATIENT 3

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	72	50	10	0·5	0·25	0·48
HOUR 1	110	50	10	0·35	0·2	1·25
HOUR 2	80	50	10	0·4	0·4	1·6
HOUR 3	100	50	10	0·4	0·4	2·0
HOUR 4	195	50	10	0·5	0·15	1·17
HOUR 5	176	50	10	0·6	0·2	1·41
HOUR 6	75	50	10	0·5	0·15	0·45

HISTAMINE STANDARD — 0·1 µg. / ml.

Histamine by mouth.

OUTPUT OF FREE HISTAMINE IN URINE AFTER 2 GMS. OF HISTAMINE BY MOUTH

(AFTER 4-5 DAYS OF SULPHASUCCIDINE)

PATIENT 3

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	62	50	10	0·5	0·15	0·37
HOUR 1	102	50	10	0·2	0·3	3·06
HOUR 2	26	26	10	0·06	0·4	6·6
HOUR 3	24	24	10	0·075	0·5	6·6
HOUR 4	32	32	10	0·1	0·3	3·0
HOUR 5	36	36	10	0·06	0·4	6·5
HOUR 6	48	48	10	0·5	0·2	4·0

HISTAMINE STANDARD — 0·1 µg. / ml.

Patient 3.

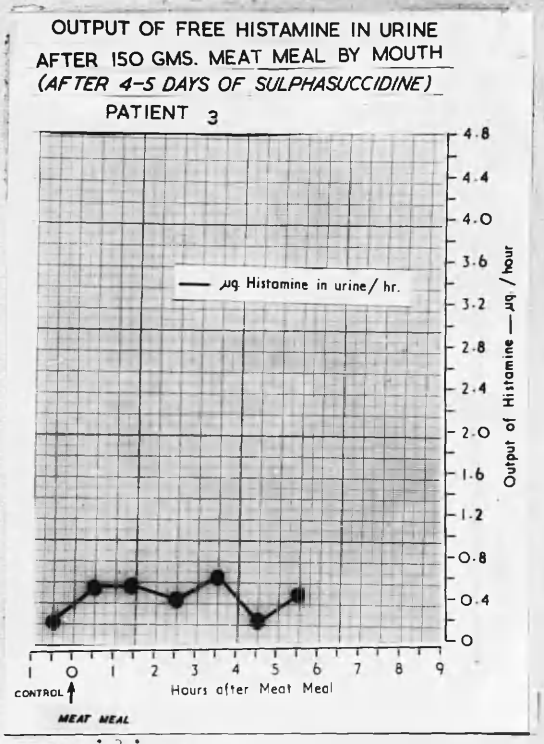
Meat meal by mouth after sulphasuccidine.

Fig.72

OUTPUT OF FREE HISTAMINE IN URINE
AFTER 150 GMS. MEAT MEAL BY MOUTH
(AFTER 4-5 DAYS OF SULPHASUCCIDINE)
PATIENT 3

SPECIMEN	TOTAL URINE VOL. ml	URINE VOL ON DECALSO COLUMN ml	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL UNKNOWN ml	BIO-ASSAY HISTAMINE ST. SOLN ml	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	35	35	10	0·5	0·1	0·13
HOUR 1	225	50	10	0·5	0·06	0·55
HOUR 2	94	50	10	0·5	0·15	0·56
HOUR 3	110	25	10	0·5	0·05	0·44
HOUR 4	66	50	10	0·4	0·2	0·66
HOUR 5	58	50	10	0·5	0·1	0·23
HOUR 6	250	50	10	0·5	0·05	0·5

HISTAMINE STANDARD — 0·1 µg. / ml.



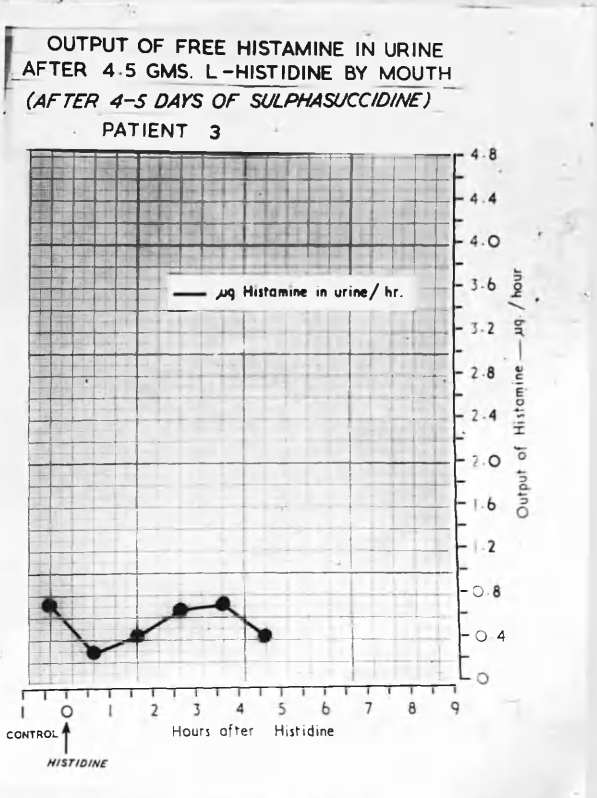
L-Histidine by mouth after sulphasuccidine.

Fig.73

OUTPUT OF FREE HISTAMINE IN URINE
AFTER 4·5 GMS. L-HISTIDINE BY MOUTH
(AFTER 4-5 DAYS OF SULPHASUCCIDINE)
PATIENT 3

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	355	50	10	0·5	0·05	0·71
HOUR 1	115	50	10	0·4	0·05	0·28
HOUR 2	36	36	10	0·35	0·15	0·43
HOUR 3	37	37	10	0·45	0·3	0·66
HOUR 4	60	50	10	0·5	0·3	0·72
HOUR 5	105	50	10	0·35	0·075	0·43
HOUR 6						

HISTAMINE STANDARD — 0·1 µg. / ml.



Patient 4.

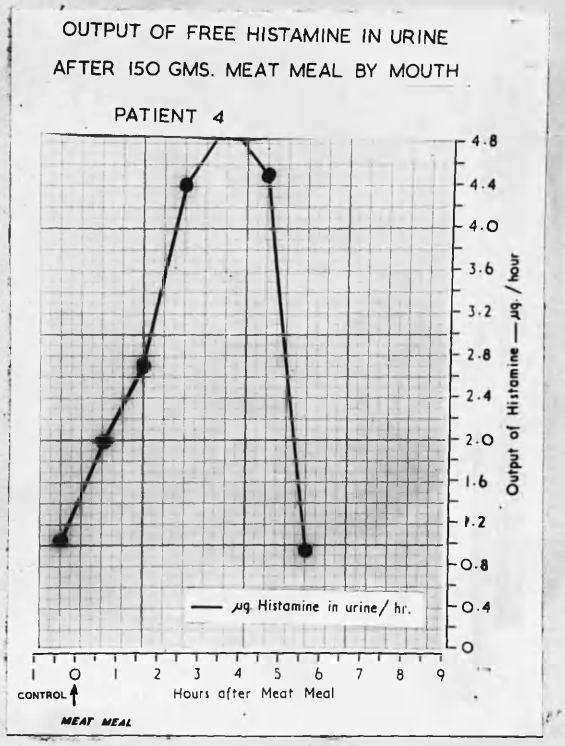
Meat meal by mouth.

Fig.74

OUTPUT OF FREE HISTAMINE IN URINE
AFTER 150 GMS. MEAT MEAL BY MOUTH
PATIENT 4

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	130	50	5	0·5	0·04	1·04
HOUR 1	120	50	5	0·3	0·05	1·94
HOUR 2	135	50	5	0·25	0·05	2·70
HOUR 3	110	50	5	0·25	0·10	4·4
HOUR 4	125	50	5	0·25	0·10	5·0
HOUR 5	180	50	5	0·3	0·075	4·5
HOUR 6	75	50	5	0·3	0·04	0·97

HISTAMINE STANDARD — 1·0 µg./ml.



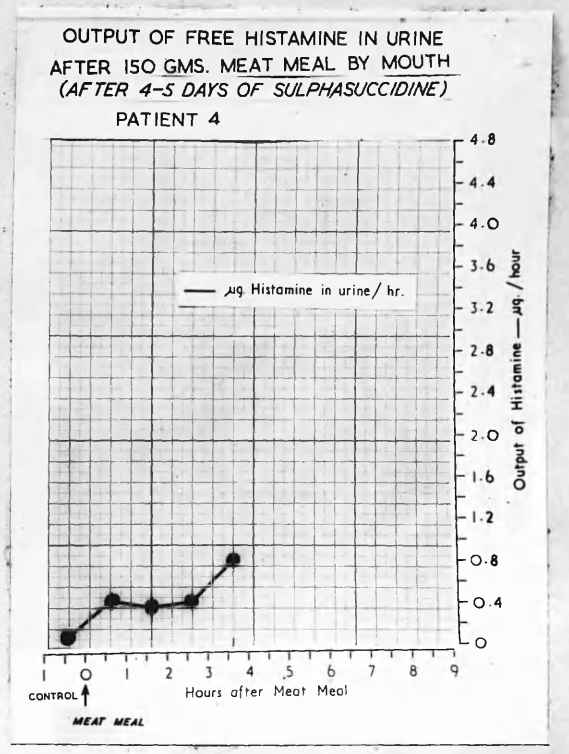
Meat meal by mouth after sulphasuccidine.

Fig.75

OUTPUT OF FREE HISTAMINE IN URINE
AFTER 150 GMS. MEAT MEAL BY MOUTH
(AFTER 4-5 DAYS OF SULPHASUCCIDINE)
PATIENT 4

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	40	40	10	0·4	0·05	0·125
HOUR 1	137	50	10	0·3	0·05	0·46
HOUR 2	200	50	10	0·5	0·05	0·40
HOUR 3	230	50	10	0·45	0·05	0·46
HOUR 4	270	50	10	0·3	0·05	0·86
HOUR 5						
HOUR 6						

HISTAMINE STANDARD — 0·1 µg. / ml.



Patient 4.

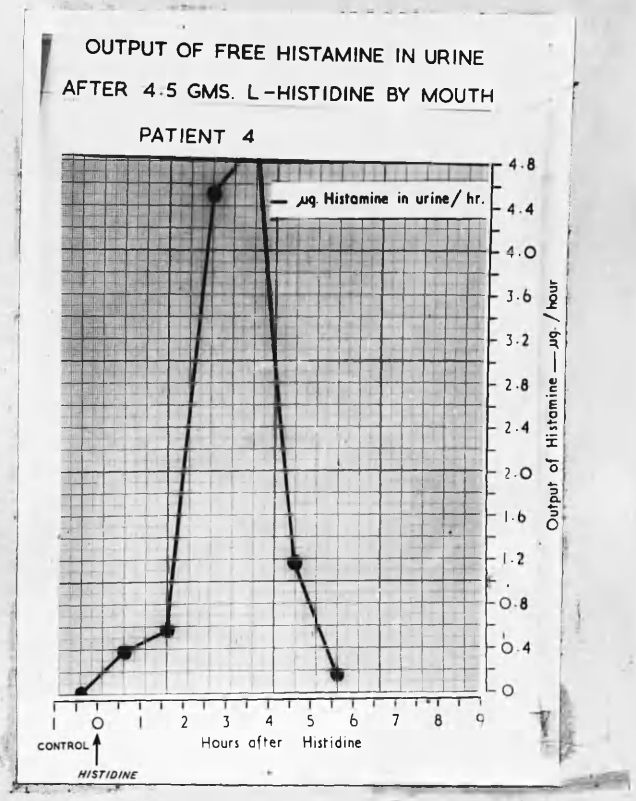
L-Histidine by mouth.

Fig.76

OUTPUT OF FREE HISTAMINE IN URINE
AFTER 4.5 GMS. L-HISTIDINE BY MOUTH
PATIENT 4

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	52	52	5	0·5	No Contraction	0
HOUR 1	115	50	5	0·6	0·02	0·38
HOUR 2	140	50	5	0·6	0·025	0·56
HOUR 3	300	50	5	0·4	0·06	4·5
HOUR 4	125	50	5	0·25	0·1	5·0
HOUR 5	115	50	5	0·3	0·03	1·15
HOUR 6	130	50	5	0·4	0·05	0·16

HISTAMINE STANDARD — 1.0 µg./ml.



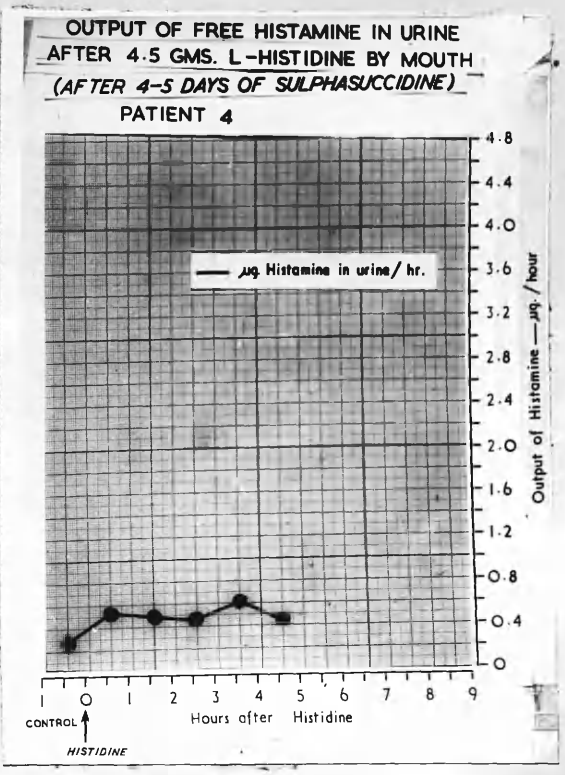
L-Histidine by mouth after sulphasuccidine.

Fig. 77

OUTPUT OF FREE HISTAMINE IN URINE
AFTER 4.5 GMS. L-HISTIDINE BY MOUTH
(AFTER 4-5 DAYS OF SULPHASUCCIDINE)
PATIENT 4

SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.
CONTROL HOUR	196	50	10	0·5	0·035	0·27
HOUR 1	440	50	10	0·4	0·025	0·53
HOUR 2	100	50	10	0·4	0·1	0·50
HOUR 3	290	50	10	0·5	0·04	0·46
HOUR 4	255	50	10	0·5	0·06	0·61
HOUR 5	28	28	10	0·35	0·15	0·43
HOUR 6						

HISTAMINE STANDARD — 0.1 µg./ml.



COMMENTARY ON RESULTS.

From a study of the results illustrated in Figs. 56 - 77 it will be seen that a 150 gm. meat meal and 4.5 g. of L-histidine each produced an augmentation of urinary histamine in all 4 patients. It can also be seen that the level to which the urinary histamine rose was roughly equivalent in both types of test. Since the concentration of L-histidine in the meat was taken arbitrarily as 3%, no closer correlation was to be expected.

The effect of sterilisation of the gut by sulphasuccidine on the output of histamine, is clearly shown in these tests. A substantial rise in urinary histamine no longer occurs after meat or histidine and the lower values obtained under these conditions are highly significant when analysed statistically.

These experiments support the concept that the decarboxylation of L-histidine by intestinal bacteria plays a major role in the augmentation of urinary histamine which is produced by a meal of meat. This process is a relatively simple one chemically and occurs rapidly, almost all the conversion to histamine being completed within 4 hours. (Waton, 1956). This fits in well with the maximal output of free histamine in the urine following a meat meal by mouth and the precipitous rise in the second hour which follows /

/follows entry of meat into the small bowel.

Even if histidine is rapidly decarboxylated to histamine is there any proof that it would be absorbed across the small bowel mucosa?

In one patient a massive dose of histamine was given by mouth (2 gms.) and the large increase in urinary histamine has been recorded in Fig. 73. In the study which follows, histamine absorption has been studied in more detail.

CONCLUSIONS.

(1) The output of histamine in the urine which follows a meat meal is parallelled when its approximate L-histidine content is given by mouth.

(2) Sterilisation of the gut significantly reduces the urinary histamine response to meat and L-histidine.

(3) It is concluded that decarboxylation of L-histidine in meat plays a considerable part in the increased output of histamine in the urine which follows a meal of meat.

CHAPTER X.

THE ABSORPTION OF HISTAMINE FROM THE ALIMENTARY LUMEN.

Evidence was presented in the previous chapter that decarboxylation of histidine to histamine in the intestinal lumen played an important role in augmenting free urinary histamine. It will be remembered that Anrep, in 1944, had noticed a difference in histamine excretion, depending on the mode of administration. When given subcutaneously, it was excreted free in the urine. After oral administration, it was excreted in the conjugated form (acetyl histamine) though traces of free histamine were also noticed. Anrep's observations were amply confirmed in man, (Adam, 1950: Adam, Hunter and Kinnear, 1950), rats (Wilson, 1954), and in dogs (Livingstone and Code). These authors were able to use more sensitive methods for measuring the free histamine output and Livingstone and Code recorded peak outputs of 15 ug/hour after administering histamine to dogs (3.0 mg/kg/hour).

In previous chapters, no evidence could be found that the occurrence of free histamine in the urine was related to gastric secretion. On the contrary, when meat was placed in the small bowel peaks of free histamine in the urine of 2 ug. or more occurred 2 hours later quite unaccompanied by significant acid secretion. Yet intravenous infusions of histamine producing a similar level of urinary histamine stimulated these gastric pouches to secrete acid. /

/acid.

The conclusion was reached that if the meat meal effects a rise in urinary free histamine by raising the blood histamine, then this rise in the blood must be in a form unable to stimulate the parietal cell. If the formation of free histamine in the intestinal lumen is indeed the source of the urinary histamine, then it must become bound in some way before reaching the general circulation. The present absorption studies were carried out to see if there was any evidence that histamine could be absorbed from the intestine in the bound form and excreted in the urine free.

OBJECTIVE OF EXPERIMENTS.

To determine if small doses of histamine, introduced into the small bowel, can produce elevations of urinary histamine without stimulating the parietal cell.

MATERIAL AND METHODS.

ANIMAL PREPARATION. Four female mongrel dogs of 18-20 kilo body weight were used for this study. Each dog had a denervated gastric pouch of Heidenhain type and Thomas cannulae were placed in the upper jejunum of each, 8" beyond the ligament of Treitz. A diagram of this animal preparation was illustrated previously in Fig. 37, page 65.

PLAN OF EXPERIMENT. The dogs were catheterised and /

/and hourly collections of acid and urine made in the usual way. For this study urine output was maintained by jejunal infusion through the Thomas cannula and this route was also used for administration of the histamine acid phosphate.

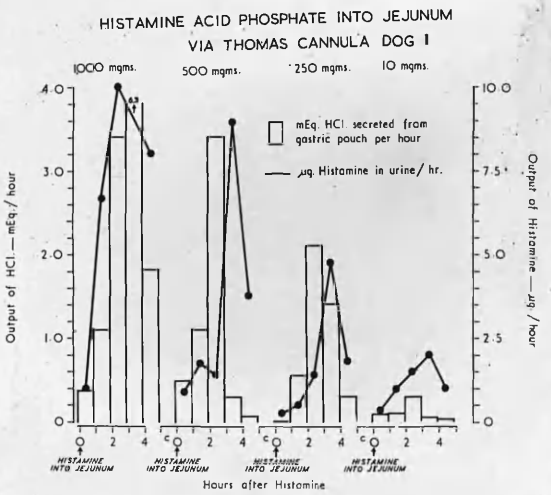
HISTAMINE ACID PHOSPHATE.

In each dog a very large dose of histamine acid phosphate (1.0 gm.) was first administered, to demonstrate unequivocally that histamine absorption occurred as measured by acid response and increase of free histamine in the urine. The dose was then reduced in stages, until one was found which produced an elevation of urinary histamine of equivalent magnitude to that obtained with a meat meal and its effect on the parietal cells noted. Since the dogs closely approximated in weight, dosage was in absolute values of histamine acid phosphate. In one dog a test was repeated after 4 days of sulphasuccidine administration (4 gms./day).

RESULTS. / In the four pages which follow, the output of urinary histamine and the acid response in the Heidenhain pouch is shown for four dogs with different doses of histamine acid phosphate introduced into the jejunum.

RESULTS. Dog 1 Fig. 78

The response of gastric pouches and the output of free histamine in the urine following various doses of histamine acid phosphate introduced into the jejunum.



HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG I 1000 mgms.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	18	18	5	0.2	0.2	1.0	2.8	0.18
	HOUR 1	8	8	"	0.03	0.2	6.65	11.0	1.1
	HOUR 2	0	0	"	—	—	—	22.5	3.4
	HOUR 3	25	25	50	0.2	0.2	10.0	43.5	6.3
	HOUR 4	350	50	5	0.026	0.2	8.0	15.0	1.81
	HOUR 5								
	HOUR 6								

HISTAMINE STANDARD — 0.2 µg./ml.

HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG I 500 mgms.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	75	50	5	0.1	0.12	0.9	6.1	0.5
	HOUR 1	35	20	"	0.2	0.4	1.75	10.2	1.1
	HOUR 2	40	40	"	0.25	0.7	1.40	26.4	3.4
	HOUR 3	210	50	"	0.1	0.42	8.93	2.8	0.3
	HOUR 4	125	50	"	0.1	0.3	3.75	1.2	0.07
	HOUR 5								
	HOUR 6								

HISTAMINE STANDARD — 0.1 µg./ml.

HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG I 250 mgms.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	9	9	5	1.0	0.05	0.25	0.2	0.005
	HOUR 1	4	4	"	0.5	0.05	0.5	6.5	0.55
	HOUR 2	30	30	"	0.35	0.1	1.4	15.1	2.1
	HOUR 3	95	50	"	0.4	0.2	4.75	9.9	1.4
	HOUR 4	21	21	"	0.4	0.15	1.8	2.5	0.3
	HOUR 5								
	HOUR 6								

HISTAMINE STANDARD — 1.0 µg./ml.

HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG I 10 mgms.

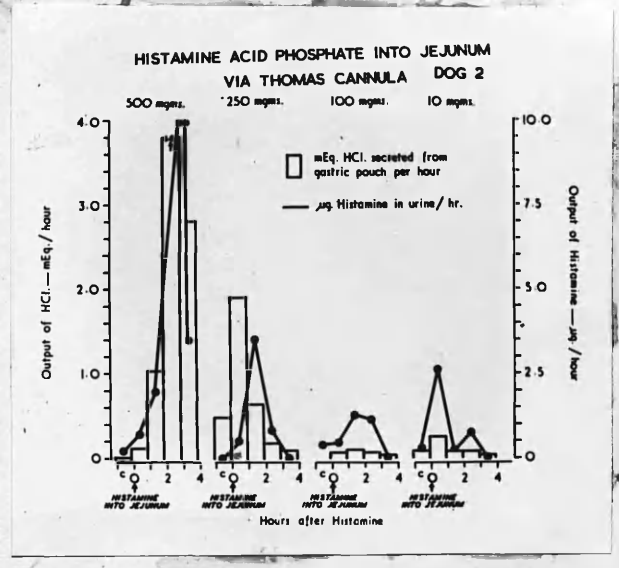
EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	70	50	5	0.2	0.05	0.35	1.6	0.09
	HOUR 1	4	4	"	0.2	0.2	1.0	2.6	0.1
	HOUR 2	27	ALL	"	0.2	0.3	1.5	2.3	0.3
	HOUR 3	18	"	"	0.2	0.4	2.0	0.4	0.05
	HOUR 4	17	"	"	0.2	0.2	1.0	1.0	0.03
	HOUR 5								
	HOUR 6								

HISTAMINE STANDARD — 0.2 µg./ml.

Dog 2

Fig. 79

The response of gastric pouches and the output of free histamine in the urine following various doses of histamine acid phosphate introduced into the jejunum.



HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG 2 500 mgms.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	10	ALL	5	0.4	0.1	0.25	1.7	0.01
	HOUR 1	5	"	"	0.2	0.15	0.75	4.1	0.13
	HOUR 2	3	"	"	0.2	0.4	2.0	9.0	1.04
	HOUR 3	18	"	"	0.04	0.6	15.0	26.5	4.6
	HOUR 4	3	"	"	0.1	0.35	3.5	17.0	2.8
	HOUR 5								
	HOUR 6								

HISTAMINE STANDARD — 0.2 µg./ml.

HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG 2 250 mgms.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	10	10	5	0.4	No Control	—	6.6	0.49
	HOUR 1	29	29	"	0.2	0.1	0.5	14.1	1.9
	HOUR 2	200	50	"	0.4	0.35	3.50	5.4	0.64
	HOUR 3	165	50	"	0.4	0.1	0.82	1.6	0.18
	HOUR 4	95	50	"	0.4	No Control	—	1.8	0.1
	HOUR 5								
	HOUR 6								

HISTAMINE STANDARD — 0.2 µg./ml

HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG 2 100 mgms.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	6	6	5	0.1	0.08	0.4	1.1	—
	HOUR 1	60	50	"	0.05	0.04	0.48	2.5	0.07
	HOUR 2	127	50	"	0.05	0.05	1.27	2.3	0.1
	HOUR 3	76	50	"	0.1	0.15	1.14	2.6	0.07
	HOUR 4	48	48	"	0.2	No Control	—	1.8	0.04
	HOUR 5								
	HOUR 6								

HISTAMINE STANDARD — 0.1 µg./ml.

HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG 2 10 mgms.

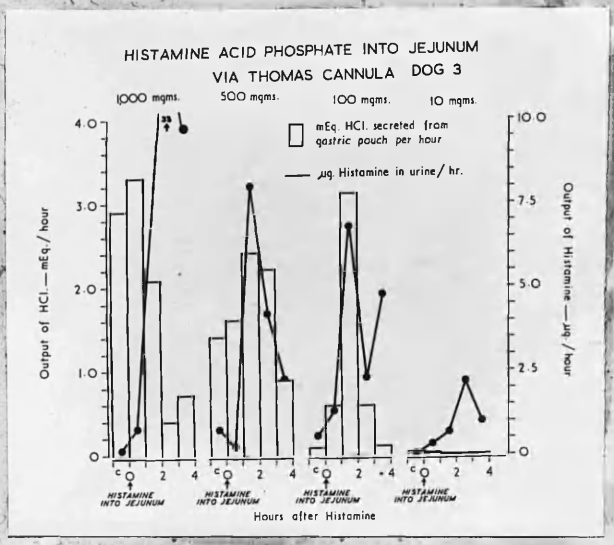
EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	30	30	5	0.5	0.03	0.3	3.2	0.09
	HOUR 1	100	50	"	0.15	0.04	2.6	4.6	0.26
	HOUR 2	175	50	"	0.4	0.05	0.22	1.3	0.08
	HOUR 3	75	50	"	0.5	0.05	0.75	1.8	0.08
	HOUR 4	17	17	"	0.6	No Control	—	1.3	0.04
	HOUR 5								
	HOUR 6								

HISTAMINE STANDARD — 1.0 µg./ml

Dog 3

Fig. 80

The response of gastric pouches and the output of free histamine in the urine following various doses of histamine acid phosphate introduced into the jejunum.



HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG 3 1000 mgms.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	55	50	5	0.6	0.1	0.16	22.5	2.9
	HOUR 1	5	5	5	0.25	0.2	0.8	25	3.3
	HOUR 2	0	0	0	–	–	–	14.5	2.08
	HOUR 3	30	30	50	0.06	0.2	33.0	4.0	0.4
	HOUR 4	385	50	5	0.3	0.4	10.07	1.5	0.72
	HOUR 5								
	HOUR 6								

HISTAMINE STANDARD — 0.2 µg/ml

HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG 3 500 mgms.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	35	35	5	0.1	0.075	0.75	14.0	1.4
	HOUR 1	60	50	"	0.1	0.04	0.24	21.7	1.6
	HOUR 2	60	50	"	0.075	1.0	7.98	21.7	2.4
	HOUR 3	240	50	"	0.2	0.35	4.2	16.6	2.2
	HOUR 4	225	50	"	0.25	0.25	2.25	6.8	0.88
	HOUR 5								
	HOUR 6								

HISTAMINE STANDARD — 0.1 µg/ml

HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG 3 100 mgms.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	8.0	8	5	0.1	0.11	0.55	6.4	0.08
	HOUR 1	6.0	6	-	0.05	0.13	1.3	10.2	0.58
	HOUR 2	104	50	"	0.05	0.325	6.76	30.8	3.1
	HOUR 3	230	50	"	0.05	0.05	2.3	10.0	0.58
	HOUR 4	175	50	"	0.03	0.08	4.75	3.0	0.10
	HOUR 5								
	HOUR 6								

HISTAMINE STANDARD — 0.1 µg/ml

HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG 3 10 mgms.

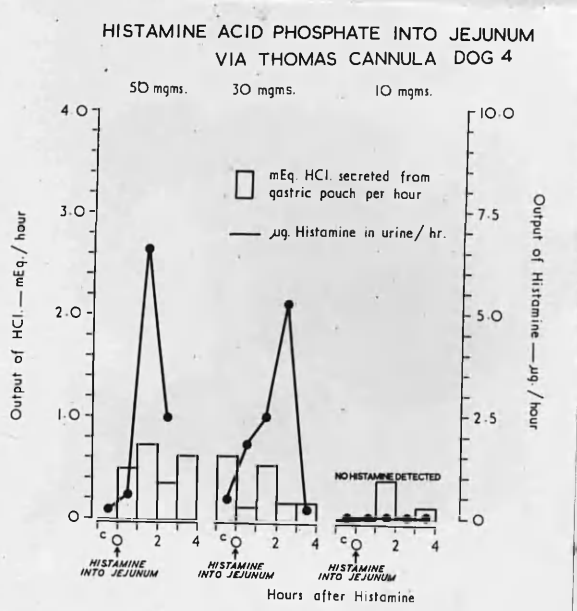
EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	128	50	5	0.4	No Control	–	1.2	0.04
	HOUR 1	150	"	"	0.1	0.01	0.3	2.2	0.02
	HOUR 2	112	"	"	0.1	0.03	0.67	3.1	–
	HOUR 3	121	"	"	0.1	0.09	2.18	2.6	–
	HOUR 4	103	"	"	0.15	0.07	0.98	2.5	–
	HOUR 5								
	HOUR 6								

HISTAMINE STANDARD — 0.2 µg/ml

Dog 4

Fig. 81

The response of gastric pouches and the output of free histamine in the urine following various doses of histamine acid phosphate introduced into the jejunum.



HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG 4 50 mgms.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	22	22	5	1.0	0.05	0.25	2.5	–
	HOUR 1	7	7	"	0.4	0.05	0.62	7.5	0.5
	HOUR 2	50	50	"	0.15	0.2	6.6	7.0	0.75
	HOUR 3	250	50	"	0.4	0.04	2.5	3.8	0.35
	HOUR 4	95	50	SPECIMEN	LOST			2.2	0.65
	HOUR 5								
	HOUR 6			HISTAMINE STANDARD — 1.0 µg./ml.					

HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG 4 30 mgms.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	50	50	5	0.5	0.05	0.5	7.0	0.62
	HOUR 1	225	50	"	0.55	0.045	1.84	13.1	0.12
	HOUR 2	250	50	"	0.5	0.05	2.5	6.0	0.55
	HOUR 3	175	50	"	0.5	0.15	5.25	4.25	0.16
	HOUR 4	30	30	"	0.6	0.03	0.25	4.0	0.16
	HOUR 5								
	HOUR 6			HISTAMINE STANDARD — 1.0 µg./ml.					

HISTAMINE ACID PHOSPHATE INTO JEJUNUM
VIA THOMAS CANNULA

DOG 4 10 mgms.

EXP.	SPECIMEN	TOTAL URINE VOL. ml.	URINE VOL. ON DECALSO COLUMN ml.	SALINE VOL. USED TO TAKE UP HISTAMINE EXTRACTED ml.	BIO-ASSAY VOL. UNKNOWN ml.	BIO-ASSAY HISTAMINE ST. SOLN. ml.	HOURLY URINARY HISTAMINE µg.	VOL. GASTRIC JUICE SECRETED ml./hr.	mEq. HCl. per hr.
	CONTROL HOUR	17	17	5	0.6	No	–	0.5	–
	HOUR 1	88	50	"	0.6	CONTR	–	0.5	–
	HOUR 2	100	50	"	0.6	AGAINST	–	9.7	0.38
	HOUR 3	62	50	"	0.6	0.045	–	1.2	0.02
	HOUR 4	55	50	"	0.6		–	4.3	0.11
	HOUR 5								
	HOUR 6			HISTAMINE STANDARD — 1.0 µg./ml.					

COMMENTARY ON RESULTS.

In all 4 dogs when more than 100 mg. of histamine acid phosphate was introduced into the jejunum, absorption of free histamine occurred, as evidenced by stimulation of acid secretion and a rise in free histamine in the urine. Even when a very large dose (1 gm.) was given, only very small amounts appeared in the urine in the next 4 hours (25 - 40 ug.).

Thus relatively minute amounts of the original dose are absorbed in the free state. The remainder is either destroyed in the intestinal lumen or converted to acetyl histamine before absorption. The acetyl histamine excretion in the urine was not measured in these studies. However, this type of histamine excretion in the urine usually only starts to rise significantly 4-5 hours after oral histamine (Anrep, 1944: Livingstone and Code, 1955), while the present studies terminated after 4 hours.

Unlike acetyl histamine, the free histamine appears immediately in the urine and its excretion is virtually over by the 4th hour of most experiments, if adequate urine output is maintained.

When 10 mg. of histamine were introduced into the jejunum, an output of free histamine occurred in the urine, parallelling the peaks seen after administration of meat either orally or directly into the intestine. At this /

/this dose level 3 of the 4 dogs produced insignificant amounts of acid from their pouches. In dog 4, no histamine was detected in the urine at this dose level. In Figs. 82 and 83 the response /

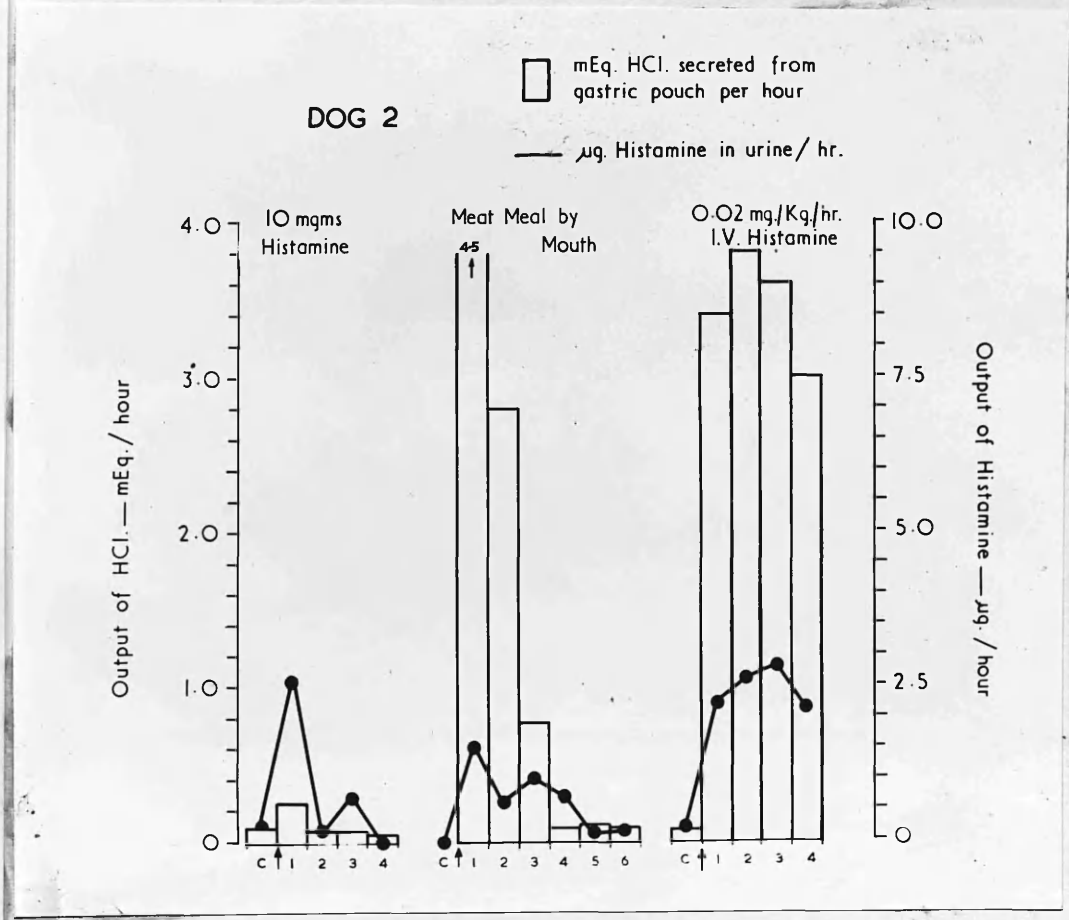


FIG. 82

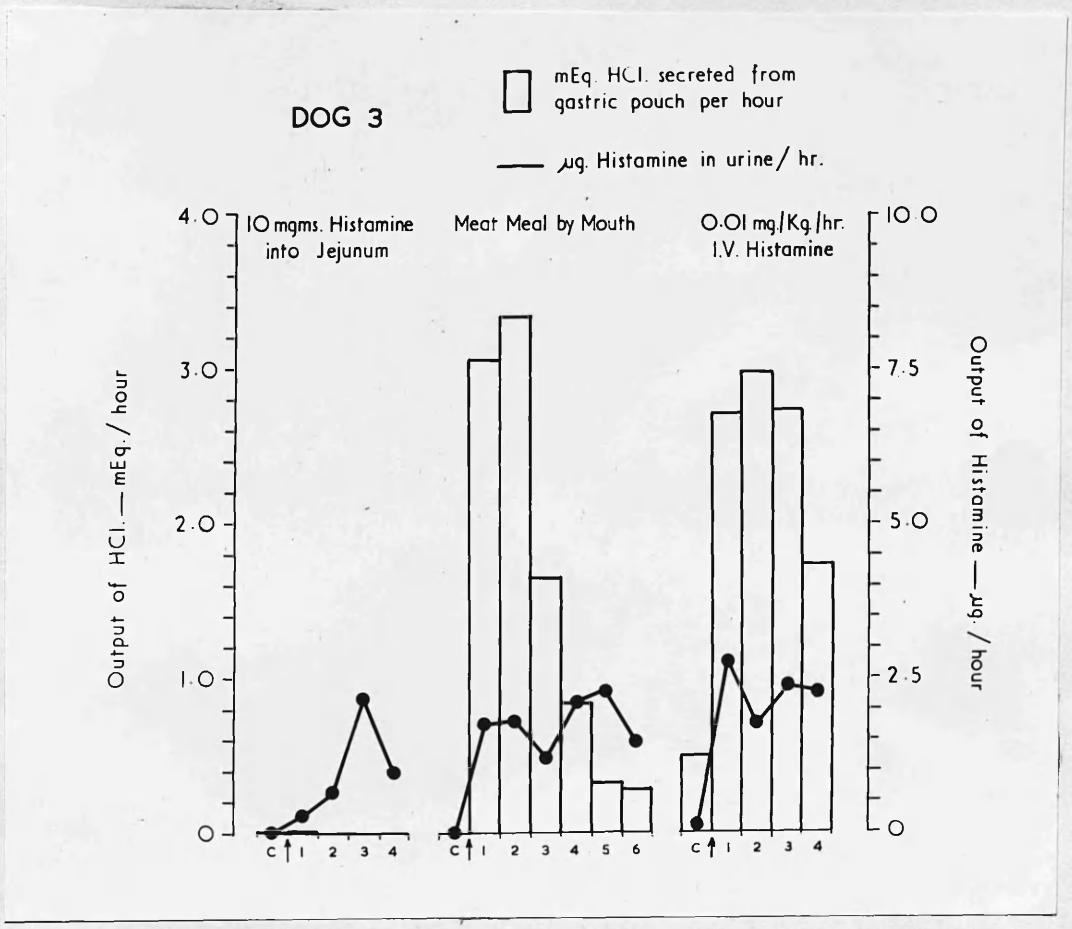

DOG 3
mEq. HCl. secreted from gastric pouch per hour
µg. Histamine in urine / hr.
10 mgms. Histamine into Jejunum
Meat Meal by Mouth
0.01 mg./Kg./hr. I.V. Histamine
Output of HCl. — mEq. / hour
Output of Histamine — µg. / hour
4.0
3.0
2.0
1.0
0
10.0
7.5
5.0
2.5
0
C 1 2 3 4
C 1 2 3 4 5 6
C 1 2 3 4

FIG. 83

/response to 10 mg. histamine acid phosphate given into the jejunum is contrasted with the result obtained by a meat meal by mouth, in the same dog. The outputs of urinary histamine are roughly equivalent in the two tests but significant acid secretion occurs only after the meat meal. When an output of urinary histamine of similar magnitude was produced by intravenous histamine the pouches strongly secreted. It is concluded that absorption of histamine from the intestinal lumen does occur. At physiological dose levels it would appear that it is absorbed in an inactive "bound form" which cannot stimulate the parietal cell.

This loosely bound histamine, if such it is, appears to be rapidly excreted in the urine in a form which is at present indistinguishable from free histamine. It must not be confused with acetyl histamine which is acetylated in the intestine by bacteria, absorbed and excreted more slowly 5 or 6 hours later as acetyl histamine.

It is suggeated that large doses of histamine break down this protective mechanism, free histamine gaining the general circulation and stimulating the gastric pouches. It may be considered that this is just a question of dosage, the smaller dose of histamine being absorbed free but failing to stimulate the parietal cell. Such an explanation is unlikely. The smallest parenteral dose of histamine /

/histamine stimulates acid secretion. Furthermore, intravenous infusions of histamine producing an equivalent overflow of free histamine into the urine, stimulated the gastric pouches while the 10 mg. of histamine introduced into the jejunum, did not. (Figs. 82 and 83).

Although these studies suggest that physiological amounts of histamine in the intestinal lumen are inactivated, or bound, before entry into the general circulation, they give no indication of the site of this process, if it does exist.

Koessler and Hanke (1924), working with guinea pigs, came to the conclusion that histamine may be rendered pharmacologically inert in its passage through the wall of the intestine. They noted that 800 mg. guinea pigs withstood doses of 100 mg. of histamine by mouth without systemic effects, while a relatively minute dose injected outside the intestinal wall produced such effects. The liver is another possible site of such a mechanism and studies of histamine absorption before and after the formation of Eck-fistulae, may help to clarify this point.

Finally it should be recorded that in any one dog considerable variations in urinary histamine and acid output occurred for a constant dose of histamine. This was particularly noticed with the smaller doses. It must be remembered that the histamine after reaching the intestinal lumen is utilised by bacteria, much of it being converted /

/converted to acetyl histamine. If in any test, this process is delayed or reduced the amount of free histamine available in the lumen for absorption will be greatly increased.

Such an effect has been demonstrated by repeating one of these histamine tests after 4 days of sulphasuccidine administration (Fig. 83).

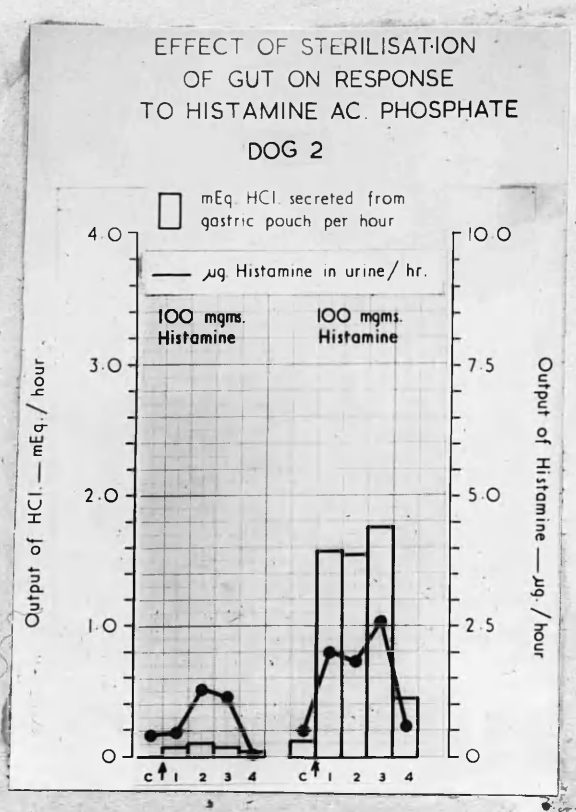
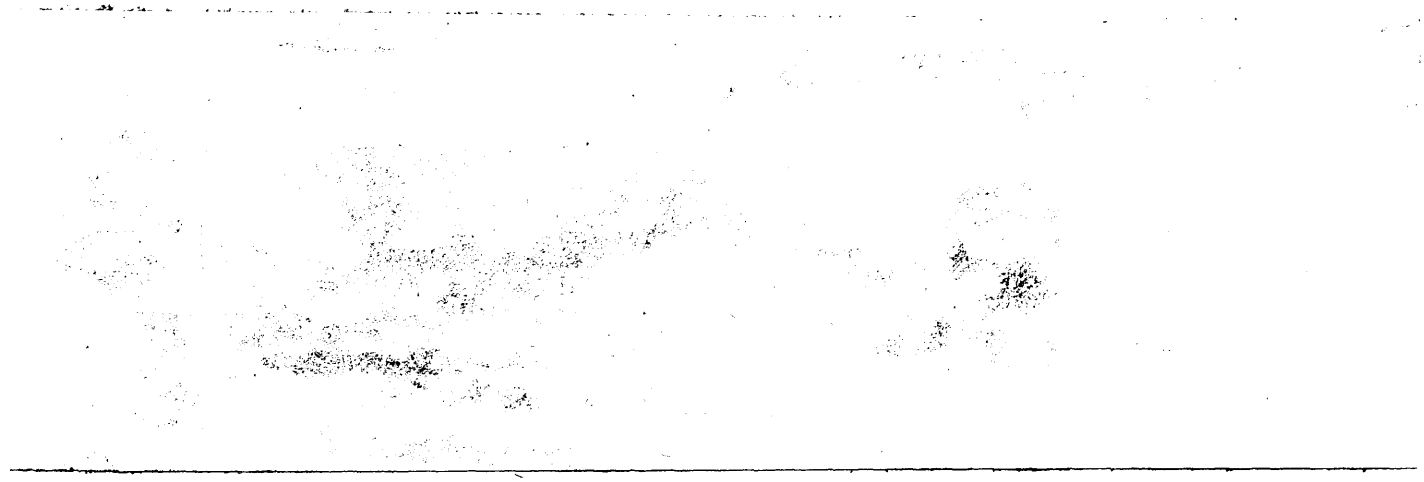


Fig. 84.

In the first test, in Fig. 84, before gut sterilisation, the histamine is absorbed in a form unable to stimulate the parietal cell. After sterilisation, the same dose, with reduced acetylation taking place, makes more free histamine available for absorption and the protective 'binding' mechanism is overwhelmed, the pouch being stimulated to produce acid.



CONCLUSIONS.

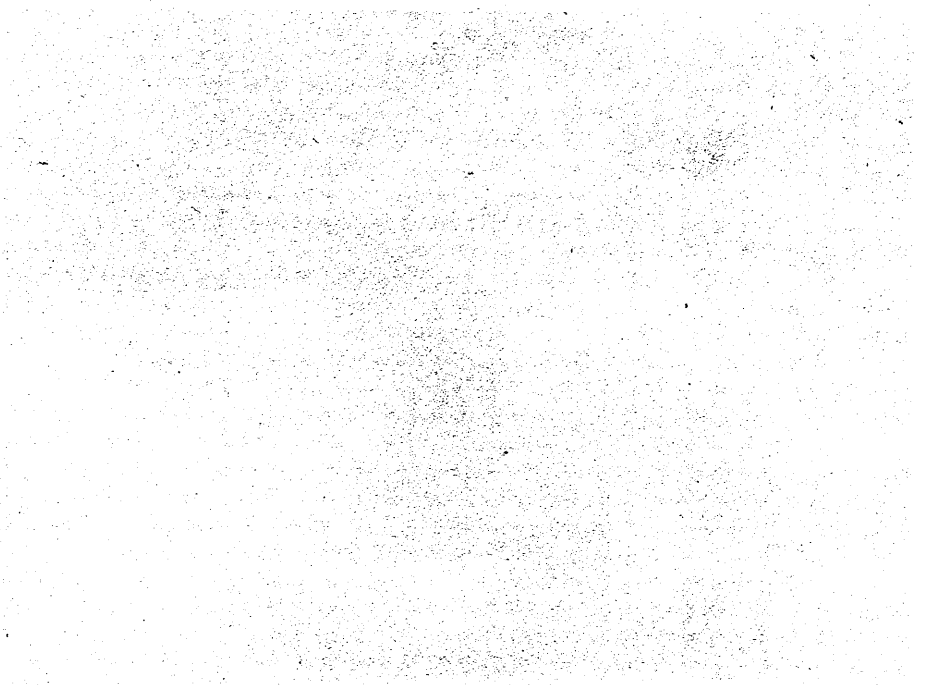
(1) When histamine acid phosphate is introduced into the small bowel lumen, minute quantities of it are absorbed and free histamine rapidly appears in the urine. This may be incompatible with the suggestion of Smith (1953) that the alimentary tract forms an important pathway for histamine excretion.

(2) Large doses of histamine (over 100 mg.) in the jejunum stimulate acid secretion as well as increase free histamine in the urine.

(3) Small doses of histamine (10 mg.) increase the free histamine in the urine at least as much as a meat meal, without /

/without stimulating acid secretion.

(4) Evidence is presented which suggests that small doses of histamine are 'bound' or inactivated before entering the general circulation.



CHAPTER XI.

FINAL CONCLUSIONS.

(1) When dogs with Heidenhain pouches take a meat meal by mouth there is a rough parallelism between output of histamine in the urine and acid secretion from the pouch. There is also a slight but definite correlation hour by hour between acid response and free urinary histamine (correlation coefficient = 0.6).

(2) The rise in urinary histamine after a meal of meat by mouth still occurs after total gastrectomy. This points against the gastric mucosa being the only source of this histamine in the urine. Since no pre-operative data were available on these dogs, no quantitative comparisons of the effect of the operation on free histamine output were possible. That one or other phase of acid secretion contributed significant quantities of free histamine to the urine was not excluded by these experiments.

(3) Vagal stimulation produced by insulin induced hypoglycaemia, in dogs with simple gastrostomies, greatly augments acid secretion without affecting the output of free histamine in the urine. /

/urine.

(4) Meat in an isolated stomach, in contact with the antrum, stimulates acid secretion from a denervated pouch without any rise in the urinary histamine being detected, by the method used.

When an equivalent acid response from each pouch is produced by an I.V. infusion of histamine, a considerable elevation of urinary histamine is detected by the same method.

These observations point against gastrin being histamine or a general histamine liberator. They do not exclude the possibility that gastrin releases histamine locally at the parietal cell.

(5) When meat is introduced directly into the small bowel of dogs with Heidenhain pouches, a precipitous rise in free histamine occurs in the urine, a peak being reached in the second hour. Thereafter the output falls rapidly and control levels are reached by the time the intestinal phase of acid secretion is maximal. This argues against histamine being the humoral agent concerned in the intestinal phase of acid secretion. When a bread and milk meal was introduced into the jejunum no such rise in urinary histamine occurred. /

/occurred.

(6) The output of histamine in the urine which follows a meat meal is paralleled when its approximate content of L-histidine is given by mouth. Sterilisation of the gut significantly reduces the urinary histamine response to these substances. It is concluded that the decarboxylation of L-histidine to histamine plays a considerable part in the increased output of histamine in the urine which follows a meal of meat.

(7) Such a concept requires that histamine be absorbed from the intestinal lumen. Furthermore, introduction of meat into the small bowel produced a rise in urinary histamine without stimulating the gastric pouch, suggesting that it was absorbed in an inactive form in these tests. Histamine absorption studies in dogs with Heidenhain pouches support this concept. Small doses of histamine introduced into the intestinal lumen produce levels of free histamine in the urine which parallel those seen after a meat meal, without stimulating the gastric pouches to secrete acid.

(8) It is concluded that the free histamine appearing in the urine and the acid secreted by a gastric pouch when a meat meal is ingested, are unrelated. /

/unrelated. The moderate hour by hour correlation between urinary histamine and acid secretion, observed in the first experimental study, may be explained on the basis that two quite separate processes are taking place about the same time.

In the early hours after the meal, the meat is initiating the various humoral mechanisms which stimulate the denervated gastric pouch to secrete. At the same time, L-histidine is being decarboxylated to histamine when it enters the small bowel. Absorption of small amounts of this histamine in the intestinal lumen, probably in a bound state, are reflected in a rise of free histamine in the urine.

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Trypsin. — Papsen.

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Emmelin. see also over page.

support for or against

---

vivax and Plasmodium falciparum.

It is very little and can be used as a little, causing gastric secretion. It causes a raise in body temperature.

Quinine slows the heart due to its quinidine-like action. It stimulates the central nervous system causing hyper-excitability, apprehension and delirium.

Muscarine

Muscarine is a sympathomimetic drug. It acts in a similar way to adrenaline Muscarine stimulates the heart giving increased blood pressure. It inhibits the [illegible] and inhibits the secretion of intestinal glands.

Progesterone

Progesterone is produced in the male in the adrenal cortex. Its action is to promote spermatogenesis.

In the female progesterone is produced in the adrenal cortex, the ovaries and in the placenta during pregnancy. Progesterone aids the implantation of the fertilized ovum in the wall of the uterus. It also favours continuation