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The Histamine Content of Allergic and Non-Allergic Human Nasal Mucous Membrane with Simultaneous Observations on the Eosinophils.

by

James Douglas Baxter

(J.B. Collip Fellow in Medical Research, 1951-52)

Thesis

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GENERAL INTRODUCTION

This investigation was undertaken at the suggestion of Dr. Bram Rose in order to assess what happens to the histamine content of human nasal mucous membrane during an allergic reaction.

In previous work carried out by Rose, Entin and Baxter (1951) on the histamine content of various human tissues, several high values (101 and 140 γ per gram) were obtained on antral mucous membrane from allergic individuals. The exact significance of such findings was not clear.

Buhrmester and Wenner (1936) reported that on the basis of moist weight, nasal mucous polyps and normal nasal mucous membrane showed almost identical histamine values. On the basis of dry weight, however, the polypoidal tissue contained more histamine than the normal. Their "normal" tissue was obtained from cadavers.

It is generally agreed today that at least part of the human allergic phenomena is based on the antigen-antibody reaction with a resulting release of histamine from the cells of the shock organ. If this is correct one would expect to find a decrease in the cellular histamine content of the shock organ during an allergic reaction.

The scanty evidence available to date, however, suggests that there is an increase in the histamine content of human nasal mucous membrane under such circumstances.

This thesis is concerned with my findings on the histamine

content of human nasal mucous membrane in allergic and non-allergic states and with simultaneous observations, whenever possible, on the eosinophils in the peripheral blood, nasal tissue and nasal secretions. These findings are correlated with clinical nasal signs and symptoms.

There now follows a general review of the role of histamine in anaphylaxis and allergy.

HISTORICAL SUMMARY

Review of the Investigation leading to the Discovery of the Presence of Histamine in Animal Tissue.

The formation of a toxic chemical substance during anaphylactic shock in animals was one of the first mechanisms suggested for the production of the symptoms of the reaction (Biedl and Kraus, 1909). That this chemical factor might be histamine was inferred by Dale and Laidlaw in 1910 when they pointed out the similarity between the physiologic action of histamine and the phenomena of anaphylactic shock.

In 1907 Windaus and Vogt synthesized histamine from histidine before it was isolated from either animal or plant sources. The procedure was a complicated decarboxylation of histidine. Three years later (1910) Barger and Dale isolated it from ergot.

During the next seventeen years, in spite of extensive investigation into the action of histamine in vivo and the similarity to the conditions of anaphylactic shock (Dale, 1913; Coca, 1914; Abel and Kubota, 1919) the question as to the existence of the substance as such in tissue was not proven.

In 1911, Barger and Dale isolated histamine in small amounts from intestinal mucosa, but it was argued that this might be due to contamination by intestinal contents.

In 1919, Abel and Kubota isolated histamine from dried pituitary powder. Two years later Hanke and Koessler (1920), using a different method, were unable to detect histamine in fresh pituitary glands.

The controversy as to whether histamine was present as such in animal tissues was finally settled in 1927 when Best, Dale, Dudley and Thorpe isolated the picrate salt from ox liver and lungs. In 1929, Dale and Dudley isolated the picrate salt from ox and horse spleen and a year later Thorpe (1930) isolated it from ox muscle.

Further investigation has established that histamine is present in most plant and animal tissue. In mammals, the amounts present vary considerably, not only in different species but also in the same organs of different individuals of the same species (Gaddum and Dale, 1936; Rose, Entin and Baxter, 1951). Apart from the observations of the latter investigators and the occasional odd determination, no acceptable series of values on the histamine content of normal human nasal mucous membrane could be found in the literature.

ANAPHYLAXIS AND ALLERGY

Introductory Remarks

It is difficult to ascribe with justice the first description of an anaphylactic reaction to any one individual. The exact origin of the subject is obscured in the development of immunologic studies in the latter part of the 19th and early 20th centuries. The use of repeated injections of solutions containing foreign protein into animals and man to induce or transfer immunity brought to light the hypersensitive state. By 1909, with the subject still in its infancy, Anderson and Rosenau found some two hundred papers worthy of mention when they reviewed the literature.

Since that time, in spite of an enormous accumulation of written material, the mechanism of the hypersensitive state, with its associated anaphylactic reaction is still not fully understood.

The early studies of hypersensitivity are linked with three great schools: a group in Germany headed by von Pirquet and Schick, a group in France led by Richet and an American school. Von Pirquet and Schick studied the hypersensitive state in human beings and their observations did much to clarify the subject. It was von Pirquet who coined the term "allergy", which is derived from the Greek words "allos" (change) and "ergon" (reaction) - "changed reaction".

Richet carried out most of his studies on dogs and it was he who introduced the term "anaphylaxis". This was derived by compounding the two Greek words "ana" (against) and "phylaxis" (protection) - "against protection". He thus defined the hypersensitive state as the opposite to prophylaxis.

In America most of the early work (Gay and Southard, 1908) (Anderson and Rosenau, 1909) was carried out on guinea-pigs and was concerned with what may be termed the mechanism of anaphylaxis.

Allergy and anaphylaxis are closely related. Zinsser (1939) believed that in reality they are one and the same manifestation. Some writers use the term anaphylaxis to denote the phenomenon of hypersensitivity in animals and the term allergy to denote a similar condition in humans. It is quite true that they have many factors in common. However, final judgement that they are one and the same manifestation must be withheld at the present time, until our knowledge of these conditions is greatly augmented.

Evidence for the Liberation of Histamine in Anaphylaxis and Allergy in Various Species.

We will now consider the evidence for the liberation of histamine in anaphylaxis in several experimental animals and in allergy in humans.

A. Anaphylaxis in the Guinea Pig. The dominant symptom of acute anaphylaxis in guinea pigs is bronchoconstriction (Otto, Gay and Southard, 1908)(Auer and Lewis, 1910).

In 1910, Schultz demonstrated that isolated perfused uteri from sensitized guinea pigs contracted forcefully upon addition of the sensitizing antigen. The response of the smooth muscle was identical with that given by histamine. This work was confirmed by Dale (1913).

Bartosch, Feldberg and Nagel (1932), using isolated perfused lungs of sensitized guinea pigs, showed that when antigen was added a histamine-like substance was liberated into the perfusate. This was confirmed and extended (Wachstein (1932), Daly and Schild (1934), Daly et al. (1935)), and it was shown that the active principle was inactivated by histaminase. There could be little doubt that the active principle was histamine.

Bartosch (1935) showed by experiments in which one lung was subjected to shock while the other was used as a control that the appearance of histamine in the venous perfusate was associated with a corresponding decrease in the histamine content of the "shocked" tissue.

In 1936, Ungar and Parrot suspended isolated intestine of a normal guinea pig in warm tyrode solution containing lung fragments from a guinea pig sensitized to horse serum. The addition of horse serum in amounts which, in themselves, had an insignificant stimulating effect produced, after a short latency, a strong contraction which was

attributed to the release of histamine from the lung fragments. A modification of this method was used independently by Schild (1937 and 1939) during which he found that histamine was liberated in readily estimable quantities from the isolated aorta, uterus, liver and lungs of sensitized guinea pigs when these tissues were exposed to the antigen.

Code, in 1939, showed that during anaphylactic shock in guinea pigs, the histamine content of the blood rose from three to nine times its control value. He showed that this rise was not due to the coincident anoxemia.

From these various experiments it may be concluded that in the guinea pig, histamine is liberated during anaphylactic shock and that it plays a definite role in the symptomatology of the reaction in this species.

B. Anaphylaxis in the Dog. Biedl and Kraus (1909) first described the two most outstanding features of anaphylactic shock in the dog, the fall of blood pressure and the reduced coagulability of the blood. They thought the fall in blood pressure was due to vasomotor paralysis caused by the formation of a toxic peptone-like substance.

In the following year (1910), Manwaring showed that the liver is the organ most clearly involved in shock in these animals. He found by cross-circulation methods that when a normal dog receives blood from a dog in anaphylactic shock, signs of anaphylaxis develop. If he excluded the liver from the circulation of the sensitized dogs this shock did not appear. This indicated that the fall in blood pressure which occurs during anaphylaxis in the dog was due to the liberation of depressor substances from the liver.

This work was confirmed by Voegtlin and Bernheim (1911), Denecke (1914), Weil (1917) and Weil and Eggleston (1917). Weil's experiments did not indicate the presence of toxic factors in the blood so he concluded that the fall of blood pressure in canine anaphylaxis was a secondary result of hepatic congestion.

In 1925, Manwaring et al. were unable to detect depressor substances in blood from the carotid artery of dogs in anaphylactic shock. However, in their experiments the blood from the liver did possess definite blood pressure lowering properties.

The problem was finally clarified by the studies of Dragstedt and his associates (1932-37). They demonstrated the presence of a vasomotor substance in blood taken from the inferior vena cava just above the diaphragm. The site is important because the vein in this locality carried a good deal of blood which has come directly from the liver.

Extensive investigation of the active substance by Gebauer, Fuelnegg and Dragstedt (1932) and Dragstedt and Mead (1936) allowed them to conclude that it was histamine.

In 1939, Code showed that during anaphylactic shock in dogs the concentration of histamine in the blood was increased from two to more than eighty times the control value. The maximal concentration of histamine was generally reached within ten minutes after injection of the shock dose of antigen. After this initial rise, the concentration fell rapidly, often returning to normal values in two to three hours. This explosive liberation of histamine coincided with the dramatic fall of blood pressure which is a prominant feature of canine anaphylaxis.

Jaques and Waters (1941) showed that the reduced coagulability of the blood occurring during anaphylaxis in dogs was due to the liberation of heparin from the liver. The preponderance of evidence definitely indicated that most of the histamine released during the reaction likewise comes from the liver. (Watanabe (1931), Ojers, Holmes and Dragstedt (1941)).

From these various experiments it may be concluded that in anaphylactic reactions in the dog, there are symptoms caused by the release of histamine and heparin and that these substances come primarily from the liver.

C. Anaphylaxis in the Rabbit. Severe anaphylactic shock in the rabbit is accompanied by a fall of carotid blood pressure. Auer (1911) noted the pronounced dilatation of the right side of the heart during fatal anaphylactic reactions and concluded that heart failure was the primary lethal factor.

The work of Airila (1914), Coca (1919) and Drinker and Bronfenbrenner (1924) has clearly shown that during acute anaphylaxis in the rabbit there is a pronounced increase of peripheral resistance in the pulmonary vascular bed due to contraction of the muscular coat of the pulmonary arterial tree and that this is the cause of the right-sided heart failure. Histamine injected into rabbits causes pulmonary vascular constriction (Dale and Laidlaw (1910), Cloetta and Anderes (1914) and Rocha e Silva (1940)).

Rose and Weil (1939) and Rose (1940) showed that the blood histamine falls in anaphylactic shock in the rabbit. In normal rabbit blood the major amount of histamine is present in the white cell elements (Code and Ing, 1937). In severe anaphylaxis leucopenia uniformly occurs and the physical properties of the white cells are altered (Abell and Schenck, 1938).

Dragstedt and his associated, in 1940, using perfusion experiments with lungs and blood from sensitized rabbits, demonstrated that on the addition of the antigen to the blood, fifty per cent of the white cells drop out of the circulation during their first passage through the lungs and, at the same time, the histamine content of the blood was reduced.

In 1940, Katz incubated blood from a sensitized rabbit with the specific antigen in vitro and demonstrated the liberation of histamine

from the cells into the plasma, where it was free and active. This work was confirmed by Dragstedt et al. 1940 and 1941, and by Rose in 1940.

For this experimental evidence we can conclude that, although the whole blood histamine may fall during anaphylaxis in the rabbit, the amount liberated from the white cells into the plasma would be sufficient to produce a marked constriction of the pulmonary vessels. In the rabbit it would seem that histamine produces the pronounced and often fatal symptom of the anaphylactic reaction.

D. Anaphylaxis in Mice and Rats. The great difficulty encountered in producing anaphylactic shock in mice and rats is paralleled by their marked resistance to histamine. In order to produce a fatal histamine shock in these animals, several milligrams have to be injected intravenously. It is, therefore, unlikely that effective amounts of histamine are released in anaphylaxis.

It has been noted (Gottesman and Gottesman (1928), Wymon (1929), Rose and Browne (1939)) that following adrenalectomy the resistance of these animals to histamine decreases markedly and anaphylactic shock may be produced. The predominant anaphylactic reaction in both animals is a fall in blood pressure.

Feldberg (1941) felt that the mild anaphylactic reaction in mice and rats was mainly the direct outcome of "cell injury", that the release of histamine only slightly accentuated the reaction and that their insensitivity to histamine determines their tolerance to anaphylactic shock.

E. Allergy in Humans. Evidence for the release of histamine during allergic manifestations in humans is confined to observations on the skin, nasal mucous membrane, blood and urine.

In the skin, the urticaria and wheals of an acute reaction may also be duplicated by injection of histamine (Lewis and Grant, 1924).

Haworth and MacDonald (1937) reported that the histamine content of the blood of normal individuals remained remarkably constant while that of asthmatic patients showed marked fluctuations. This finding was confirmed by Rose (1941). He showed, with patients suffering from dermagraphia and cold allergy, that during the formation of extensive wheals histamine may be liberated from the skin and appear for brief periods in increased quantities in the blood.

Katz and Cohen (1941) incubated blood from allergic patients with various allergens and showed that the allergens to which the patient was susceptible caused a release of histamine from the blood cells into the plasma. Hawkins et al. (1951) obtained similar results with fragments of lung removed from allergic patients.

Katz (1942) devised a method whereby he could test for the liberation of histamine during local allergic reactions in the skin. With patients showing a skin reaction to the intradermal injection of ragweed, he observed a sharp increase of output of histamine from the skin into which the antigen had been injected.

Serafini (1948) observed a brief rise of blood histamine when horse dander was injected intradermally in 54 places at once in a sensitized patient.

In recent studies, by means of cardiac catheterization, Rose

Rusted and Fownes (1950) were unable to show any increase in the histamine content of the blood taken from the pulmonary and femoral arteries during induced attacks of asthma.

At our present state of knowledge, it appears too soon to say much about the urinary excretion of histamine in disease. Adam et al. (1950) did find that the rate of excretion of free histamine was higher in patients suffering from urticaria than in other patients or in normal individuals. Rose et al. (1950) showed, following ACTH, that moderate to relatively enormous amounts of histamine were found in the urine of asthmatics.

The evidence on nasal mucous membrane was discussed in the General Introduction.

From the available facts, it would seem that histamine does play some role in allergy.

ANAPHYLAXIS AND ALLERGY

GENERAL DISCUSSION

From the evidence reviewed, it appears that histamine plays a role in anaphylaxis in various experimental animals and in allergy in humans.

Its degree of importance in this reaction varies from species to species. In mice and rats it plays little if any role in anaphylaxis, while in the guinea pig, dog and rabbit it appears to have a major significance. In humans it has been shown that there is, in certain allergic conditions, a disturbance in histamine metabolism.

However, it is important to realize that histamine is not the fundamental factor in anaphylactic or allergic reactions and that its liberation may be purely incidental. The fundamental etiologic factor is the damage done to the sensitized cells during the reaction. If the damaged cells happen to contain histamine, it will be liberated. It is unlikely that histamine produces the damage.

Liberation of histamine may be a dramatic and lethal factor if the quantities liberated are sufficient, and if, as in the guinea pig, the animal is sufficiently sensitive to its effects. It has been pointed out (Code, 1939) that "anaphylaxed" dogs may die hours after the increase of histamine in the blood has disappeared. Such animals, it seems likely, die as a consequence of the reaction which liberated the histamine, and not as a direct result of the latter.

Histamine is not the only substance liberated. In dogs during anaphylaxis, Jaques and Waters (1941) showed that heparin was

liberated from the liver. It is also known that acetyl choline as well as other substances are liberated (Gaddum and Dale, 1936).

In summary, the fundamental mechanism of anaphylactic and allergic reactions lies in the process producing the damage to the cells, as a consequence of which histamine and other substances are liberated.

HISTORICAL REVIEW

Physio-Pathology of Human Allergy

Lewis (1927) originated our present conception of the allergic mechanism. From the triple response of local anaphylaxis in the human skin, he concluded that the antigen-antibody reaction in sensitized cells represented a special form of cell injury which led to the release of preformed H-substance. This H-substance is regarded today as histamine.

The pharmacological action of histamine can be summed up, according to Gaddum (1951) by saying that it causes a contraction of most smooth muscles, a dilatation of capillaries and a secretion of most glands, particularly the acid-secreting glands of the stomach.

Comprehensive studies of the histopathology of human allergy are few, mainly because of the understandable scarcity of biopsy material and the disadvantages of autopsy specimens. The latter, however, afforded Huber and Koessler (1922) opportunity to make a detailed pathological study of the lung in asthma. They found the condition was characterized by varying degrees of mucous glandular and smooth muscle hypertrophy and eosinophilic infiltration.

Nasal changes in allergy have been described and illustrated in detail by Hansel (1929-30), Cameron (1935) and Eggston and Wolff (1947). They are characterized mainly by oedema, glandular hyperactivity and eosinophilic infiltration.

The similarity between the histopathological findings in allergy and the resultant of the pharmacological action of histamine is striking. The only major factor not in common is the eosinophilia.

Rawlins (1947) felt that in nasal allergy the liberation of histamine from the damaged cells probably played an important role in the pathology but that it may not be the primary factor. He felt that the physiopathology of human nasal allergy was as follows:-

1) Antigen-antibody complex causes damage to the cells of the shock organ, resulting in the liberation of histamine from the damaged cells.
2) This histamine causes:
   a. Capillary dilatation and increased capillary permeability in the shock organ, resulting in oedema.
   b. Increased glandular activity resulting in excessive nasal secretion.
3) Associated but not dependant on this, there is an eosinophilic infiltration into the mucous membrane of the nose and sinuses and also into the nasal secretions. This aspect will be discussed later.

In nasal allergy, for obvious reasons, there is no smooth muscle component involved in the reaction.

At our present state of knowledge the foregoing appears to be the most acceptable explanation of the mechanism of nasal allergy.

HISTORICAL REVIEW

Nasal Mucous Polyps

Explanatory note: The allergic patients reported in this thesis are cases of nasal allergy with polyp formation. Because of the danger of post-operative haemorrhage arising following the removal of "non-polypoidal" allergic nasal mucous membrane in ambulatory individuals, it was necessary to confine our investigation to allergic patients with nasal polyposis. In such cases it is a simple, usually uncomplicated procedure to remove sufficient biopsy material for study.

In ancient writings the term "polyp" was applied generally to all nasal tumors (Hippocrates, 460-357 B.C.; Avicenna 980-1036 A.D. - as cited by Eggston and Wolff, 1947) but during more recent times was restricted to only soft tumors of the nose (Deschamps, 1804).

The pathologists of the latter part of the 19th century - Virchow, Billroth, Weichselbaum and Hoppmann - considered nasal polyps as an isolated phenomenon not connected with infection of the sinuses. They were considered neoplastic lesions and were designated as fibromas, mxyomas, fibro-adenomas or cystic fibro-adenomas.

The modern rhinologist and pathologist realize that, although true tumors may occasionally occur in the nose, mucous polyps represent essentially an oedema and simple hypertrophy of the nasal mucosa.

As early as 1907, Yonge stressed oedema as the starting point of nasal polyps.

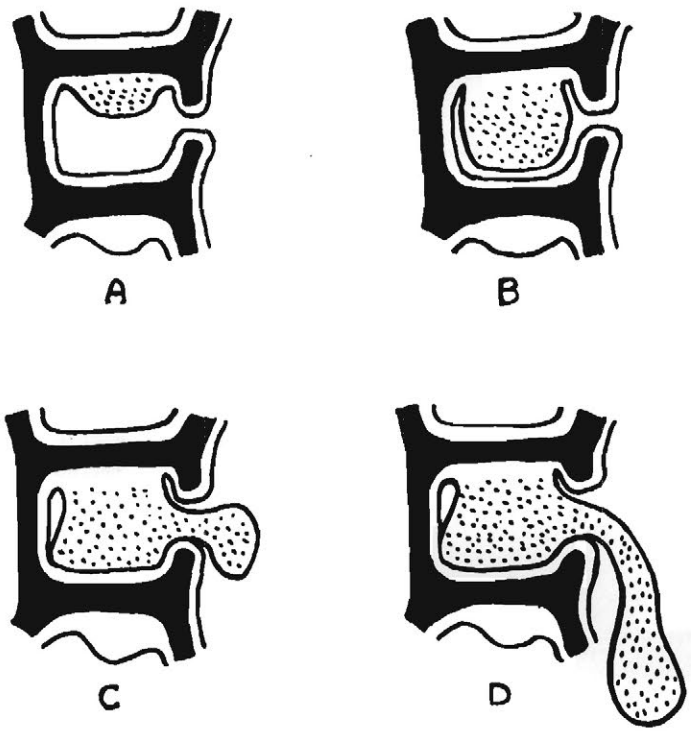
During the past fifty years, two principal schools of thought have arisen in the literature with reference to the etiology of nasal mucous polyps. These are:

1) The Allergic School

2) The Infectious School

Due primarily to the work of Kern and Schenck (1932-33, 1934, 1938) it is now generally recognized that nasal mucous polyps have an allergic basis (Hirsch, 1929; Hansel, 1936; Ungar, 1945; Morrison, 1948). They occur particularly in those types of respiratory allergy that are protracted and perennial. In such cases the persistent allergic oedema of the tunica propria in certain specific areas plus the effect of gravity leads to a prolapse of the mucous membrane with subsequent polyp formation. (See Fig. 1, page 22). Once an area of mucosa has prolapsed, its own weight will tend to hasten the formation of a stalk and, in turn, constriction of the structures in the stock exaggerating oedema in the polyp (See Fig. 2, page 23). Other factors involved in the mechanism include differences in the texture of the nasal mucosa, anatomical situations and the traction of nasal secretions.

Mucous polyps never grow from the roof or floor of the nose and rarely from the septum or inferior turbinate. They almost invariably arise from the outer nasal wall and principally from the middle meatus and the cells of the ethmoidal labyrinth. More rarely a polyp may take its origin in one of the accessory sinuses other than the ethmoidal and protrude through the natural ostium into the nose. This occurs most frequently in the maxillary sinus, and then the polyp generally passes backwards to hang into the post-nasal space (See Fig. 3, page 24).



Mode of formation of an ethmoidal mucous polyp (Diagrammatic-Morrison (I948)).

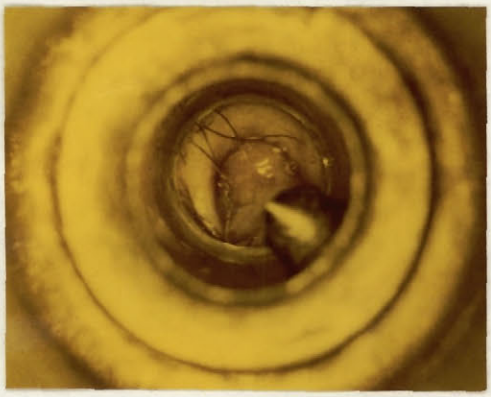
Four successive stages in the formation of a mucous polyp from an ethmoid cell, cut in vertical cross section. (A) Oedema of the mucosa of an ethmoid cell most marked on its roof. (B) Increase of the oedema of the mucosa on the roof of the cell, plus the influence of gravity, leads to a prolapse of the mucous membrane and polyp formation. The polyp is confined to the cell cavity. (C) Further increase in the size of the mass fills the cell; pressure against the margins of the ostium causes atrophy of its bony margins and the polyp begins to protrude into the nasal cavity. (D) Further action of gravity on the dependent mass causes it to hang down and grow larger and its pedicle to become more slender; the polyp protrudes freely into the nasal cavity.

FIGURE I

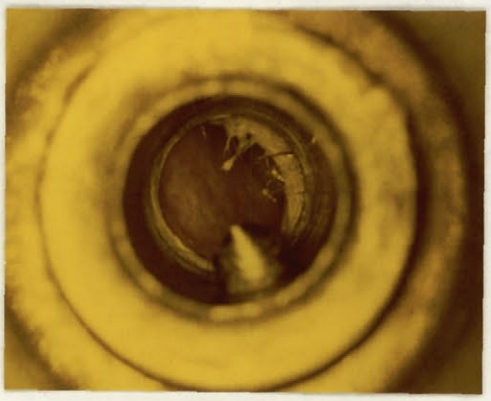
FIGURE 2

Nasal mucous polyp arising from an ethmoid cell. Note the length and constriction of the stock. The tissue in the upper portion of the photograph is the mucous membrane lining from the ethmoid cell. The specimen was in formalin for 24 hours before being photographed and has lost its characteristic "oedematous" appearance.

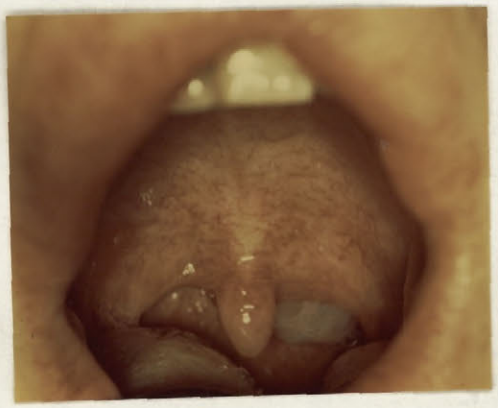
FIGURE 3



A. Nasal mucous polyp occluding the anterior nares. The mucous membrane on the exposed surface of the polyp has undergone metaplasia to stratified squamous epithelium (see Figure 5 - Page 37).



B. Nasal mucous polyp arising from the middle meatus (portion of polyp visualized through anterior nares).



C. Antro-choanal polyp presenting in the oropharynx.

Nasal polyps are rarely found before the age of twenty, and are said to be more common in males than females. They are usually bilateral but can be unilateral.

There still remains today, more especially amongst pathologists, appreciable support for the view that nasal polyps are inflammatory in origin. Ewing (1940) regarded nasal mucous polyps as one of "the purest examples of pseudo-tumor of inflammatory origin".

Eggston (1930-33) felt that polyps arose because of basic vascular changes in the nasal mucosa induced by chronic or repeated attacks of sinusitis. These insults lead to a periphlebitis or a perilymphangitis, resulting in obstruction of the return flow of interstitial tissue fluid and consequently oedema. The arterial supply remained intact.

In several ways this mechanism denotes a progressive and irreversible process and it fails to account for those cases of polyposis which regress and even disappear completely under proper management.

Many mucous polyps occur without any preceeding nasal infection. It is also noted, according to Shambaugh (1945) and Hansel (1948), that at least 70% of chronic sinus infections and 90% of chronic nasal infections can be shown to have an underlying allergic factor responsible for the chronicity. These figures may appear somewhat high, but from personal clinical experience I would tend to agree with them. Chronic nasal and sinus infections in most cases are superimposed on nasal allergy and it is usually impossible to alleviate the infection without first controlling the allergy. In view of this, it is difficult to accept the infectious theory as the primary cause.

In summary, it has been established that nasal mucous polyps are allergic in origin. There are, however, many questions concerning their formation still unanswered.

Eosinophils in Peripheral Blood, Nasal Tissue and Nasal Secretions.

The first recorded description of a leucocyte with acidophilic granules was made by Warthin Jones in 1846, but it was Ehrlick, in 1879, who named this cell the "eosinophil".

The eosinophil (Sherman, 1951) is a polymorphonuclear granulocyte of the same size as the neutrophil, having a nucleus which is usually bilobed, slightly larger and less deeply stained than the nucleus of the neutrophil. The granules of the eosinophil are characteristic, being coarse, uniformly large and ovoid, and taking a deep red stain. These cells are less motile than neutrophils, but they show phagocytosis and chemotropism of the same order as neutrophils. They are known to contain iron, oxidase, peroxidase and histamine.

In the peripheral blood it is generally agreed today (Answers to Queries, 1949) that eosinophilia is not present unless the eosinophil count is greater than 6% or the total number of eosinophils is more than 600 per cmm. A number of disorders may produce mild eosinophilia. These disorders are practically all allergic or parasitic or consist of involvement of the bone marrow. If the eosinophil determination is greater than 20%, the diagnostic field is somewhat restricted. Trichinosis is most commonly associated with such high percentages but eosinophilic leucemia, periarteritis nodosa or lymphosarcoma may show such percentages. An eosinophilia of less than 20% may indicate any one of a great number of parasitic infestations, allergic states or even bone marrow involvement.

Bizzozero (1887), Gollasch (1889), Schmidt (1891) and Wright (1898) emphasized, in cases of bronchial asthma, nasal polyps and spasmodic coryza, the finding of eosinophilic infiltrations and oedema in the tissues and the demonstration of eosinophils in the secretions. In 1895, Seiffert and Kahn illustrated by means of coloured plates the oedema and eosinophilic infiltration in nasal polyps.

Following these early observations, little importance was placed on these findings until the recognition occurred, some twenty years ago, of the role of allergy in nasal disease. Through the work of Hansel (1929, 1933-34, 1936, 1940), Finck (1927), Mullin and Ball (1928), Coates and Ersner (1930), Weille (1930), Walsh and Lindsay (1933 and 1934), Eyermann (1927), Sewall and Hunnicott (1929), Kahn and Stout (1932), Johnson and Goldstein (1932), Cowie and Jimenez (1936) and many other authors, the significance of eosinophils in nasal tissue and nasal secretions was emphasized. It is now recognized that the demonstration of eosinophils in the tissue and secretions from the nose and paranasal sinuses is good presumptive evidence of the existence of allergy (Hansel, 1933-34).

Although the exact function of the eosinophil is still unknown, it seems to be related to the removal or detoxification of foreign material and it is believed that it is part of the body defenses against invasion by heterogenous proteins; that is, allergy and parasitic infestation.

In the last few years, with the introduction of Cortisone and ACTH, great interest has been centred on the eosinophil, but it is not my intention to discuss these various observations. Note is also

made of the recent work on diurnal variations of eosinophil counts and its significance (Mann and Lehmann, 1952).

It is believed today that the eosinophil in allergic tissue is derived exclusively from bone marrow. (Opie (1904), Huber and Koessler (1922)). Opie felt that the eosinophils in connective tissue differed in no way from those of the blood. Nor does it appear that anyone has yet published evidence of eosinophil mitosis or primitive eosinophils in allergic tissue.

Some authors (Bezancon and Bernard, 1930) have expressed doubt of this exclusive bone marrow production because they felt that the numbers of eosinophils present in allergic tissue could not be accounted for except on the basis of local production. Ehrlich himself, according to Opie, suggested that, in the frog, eosinophils might be derived by a process of transformation of connective tissue cells.

The occurrence of distinct tissue eosinophilia in the absence of blood eosinophilia has proved a principal difficulty in accepting the bone marrow as the sole source of eosinophils (Cowie and Jimenez, 1936). However, Heineke and Deutschmann (1906) showed that an abundance of eosinophils may be mobilized although their percentage in the blood is within normal limits. Huber and Koessler (1922) found that tissue eosinophilia in asthma is not a uniform change, being more intense in some areas than in others. In consequence, the number of eosinophils withdrawn from the circulation to produce tissue eosinophilia is probably much less than might be anticipated.

Two types of eosinophils have been described in allergic

tissue (Huber and Koessler, 1922; Cameron, 1935), namely, polymorphonuclear, as seen in the blood, and a mononuclear form. Cameron felt that the duration of the lesion was indicated by the type of eosinophil present. In his view, acute allergic changes were associated with polymorphonuclear eosinophils whereas the mononuclear cells were a feature of chronic lesions. Huber and Koessler found that close scrutiny under high magnification resolved many mononuclear into polymorphonuclear eosinophils. They accepted a small proportion of the eosinophils as genuine mononucleated cells, but they regarded these as degenerate because these cells were no larger than healthy eosinophils and their nuclei were shrunken and pyknotic.

Experimentally, local and general eosinophilia have been produced by the injection of drugs, sera, bacterial products, pollens, foods, inhalants and histamine. In animals it has been produced by the stimulation of the sympathetic nerve endings (Hansel, 1940).

Nasal eosinophilia has been shown to be associated in some way with the state of alkalosis in the nose (Polson, 1943; Fabricant, 1945). Normally the nasal pH is on the acid side, but during acute rhinitis (infectious or allergic) there is a shift to the alkaline side and eosinophils are only present in the secretions during this latter state.

As previously mentioned, it is recognized today that the demonstration of eosinophils in the peripheral blood, nasal tissue and nasal secretions is good presumptive evidence of the existence of allergy. It is a fact, however, that they may be present in conditions other than allergic, and as such cannot be used as a

definite diagnostic indicator of allergy. In spite of extensive observations, we still know really very little about the formation and function of the "elusive eosinophil".

CLINICAL INVESTIGATIVE WORK

GENERAL INTRODUCTORY REMARKS

This work was carried out to assess what happens to the histamine content of human nasal mucous membrane during allergic manifestations in the nose. Simultaneous observations were made on the eosinophils in the peripheral blood, nasal secretions and, whenever possible, in the nasal tissue. These observations were correlated with nasal signs and symptoms.

The allergic patients studied are cases of nasal mucous polyps. These patients were observed either through the Allergy Clinic, or the Department of Otolaryngology, Royal Victoria Hospital. Many of the latter were private patients of the Attending Staff, Department of Otolaryngology, and the author is indebted to them for their co-operation.

These patients, whenever possible, were investigated thoroughly by the Allergy Department. These investigations included skin tests.

As no acceptable series of histamine determinations on normal human nasal mucous membrane could be found in the literature it was necessary to establish control values.

Through the co-operation of the Department of Otolaryngology, the author was provided the facilities to obtain sufficient normal nasal mucous membrane for histamine determinations.

METHODS

A. Histamine Determinations on Tissues

The histamine determinations on the tissues reported in this thesis were obtained by the method used in the Medical Laboratory, Royal Victoria Hospital.

The method used for the extraction of histamine from the tissue was essentially that described by Best and McHenry (1930) and the assays were performed on guinea pig ileum as described by Guggenheim and Loeffler (1916).

All the histamine values reported in this thesis are expressed as gamma per gram of wet tissue.

Histamine Values - Wet Weight versus Dry Weight of Tissue

During the recent work of Rose, Entin and Baxter (1951), but not reported in their paper (Personal Communication, Mrs. E.V. Harkness), a series of simultaneous histamine determinations were carried out on antral mucous membrane and nasal mucous polyps using both wet weight and dry weight of tissue. By wet weight we mean the weight of freshly removed tissue and by dry weight we mean the weight of tissue that has been reduced to a state of dryness by heating.

It was found that for both antral mucous membrane and nasal mucous polyps the histamine determinations on the basis of dry weight as compared to that on the basis of wet weight were consistently from three to five times greater.

In a series of such determinations on mucous polyps removed under varying degrees of nasal symptoms, the ratio (dry:wet; 3-5:1) was found to remain valid.

The significance of this finding is that a dilution factor does

not play a major role in any variations of histamine which may occur in nasal mucous polyps with varying degrees of allergic symptoms.

For this reason only wet weight histamine determinations were performed on the material reported in this thesis.

B. Eosinophils

The following methods were used for the eosinophil determinations.

i) Peripheral blood

Blood smears were prepared by the usual skin puncture method, allowed to dry and were stained with Wright's stain. Differential counts were performed. Two hundred white cells were counted per slide and the eosinophils were expressed in terms of percentage.

ii) Nasal smears

The nasal smears were prepared by either of two methods, depending upon circumstances.

If sufficient nasal secretions were present the patient was made to blow his nose and to collect the secretions on waxed paper or cellophane. If minimal secretions were present these were collected, under direct vision, on a nasal swab.

The secretions were then transferred to a clean microscopic slide and spread thinly and evenly over the surface. Any clumps or clusters of material were gently teased apart. The smear was allowed to dry and was then stained with Hansel's stain. This is a commercially prepared stain* which has been generally adopted by rhinologists in the United States and Canada for nasal smears as it has a special affinity

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* Obtainable through: Lide Laboratories Inc., 634 N. Grand Boulevard, St. Louis 3, Mo.

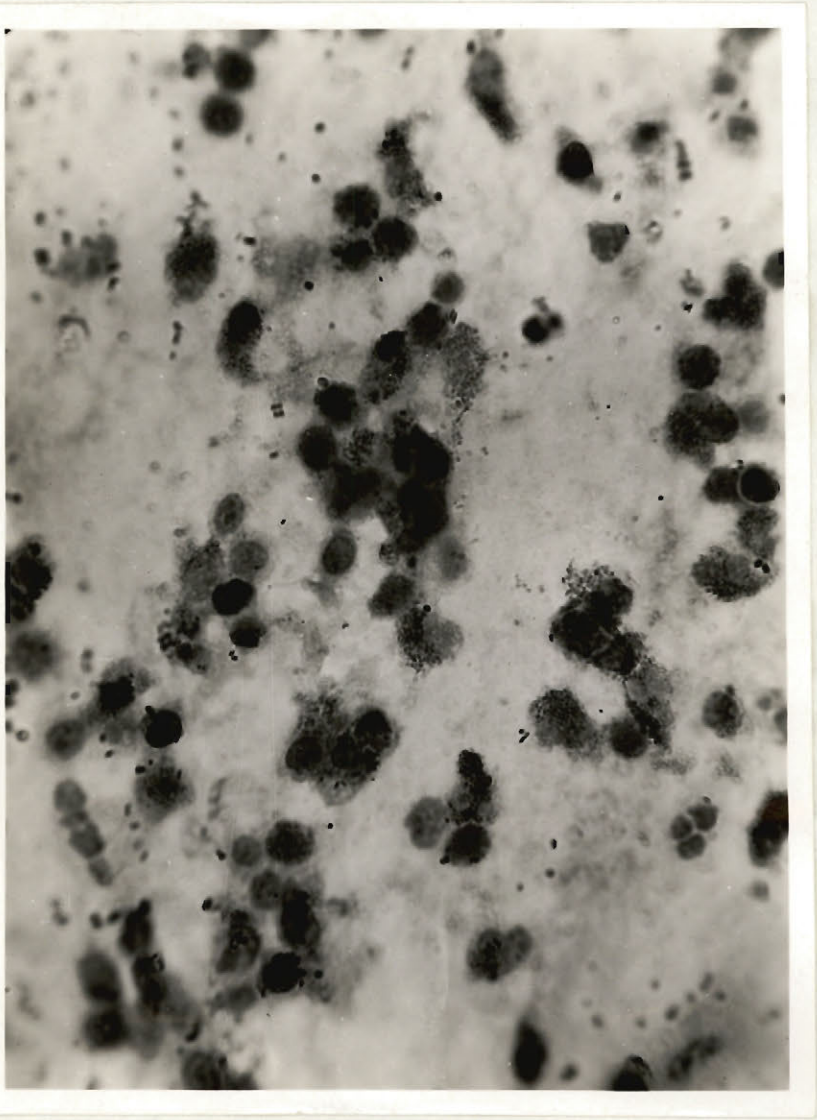


FIGURE 4

Nasal smear stained with Hansel's stain which is specific for eosinophils. Notice the prominence of the granules in the cytoplasm of the eosinophils (Magnification x 900)

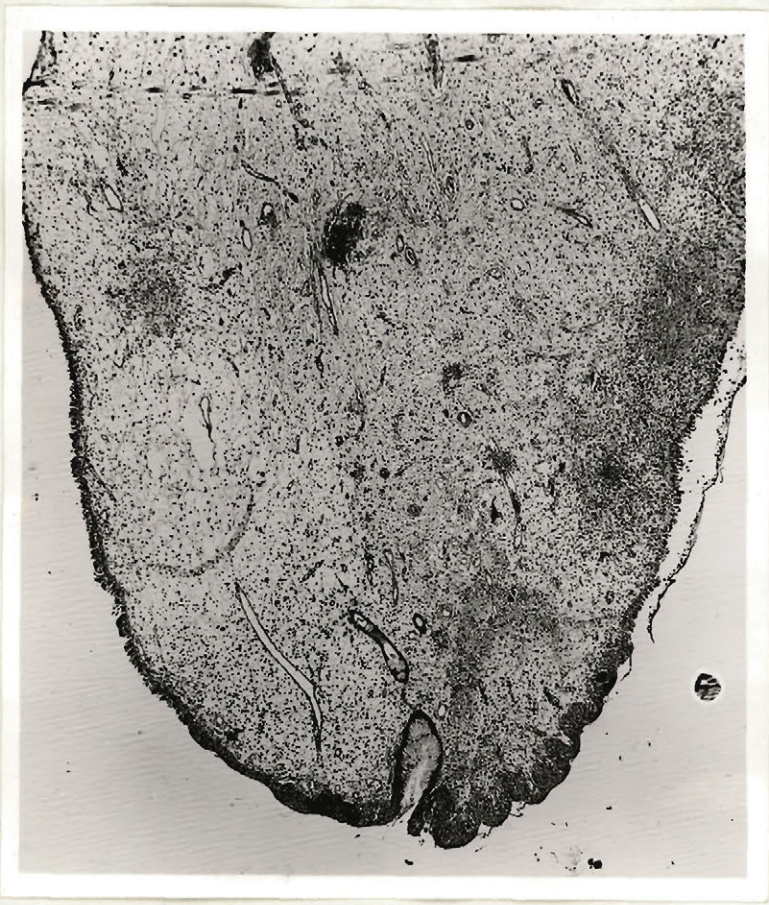


FIGURE 5

Nasal mucous polyp. Typical pathological section showing oedematous stroma and cellular content. The normal respiratory pseudo-stratified ciliated columnar epithelium in the apical portion of the polyp has undergone metaplasia to stratified squamous epithelium. This occurs whenever the membrane is exposed to repeated trauma such as in the anterior nares (Figure 2A - Page 24). In such a situation the membrane is exposed to the "blast" of the inspired air. (Magnification x 35)

for staining eosinophils (See Figure 4, Page 36).

The smear was flooded with the undiluted stain for thirty seconds, then the stain was diluted half and half with distilled water for a further thirty seconds and then the dilute stain was washed off with distilled water. The excess stain was removed by washing with alcohol and the smear was allowed to dry. It was examined under high magnification.

The interpretation of nasal smears can be somewhat complex and it is difficult to express the number of eosinophils present in terms of percentage. On examination of a smear a general impression is formed rather than an actual cell count. The results are expressed as a matter of degree depending upon whether no eosinophils are present, few are present or many are present, and degrees thereof. They were expressed as 0, +, ++ or +++ depending upon the degree present.

It is important in interpreting the nasal smears to scan the whole slide and get a general picture as the eosinophils tend to cluster and may only be present in certain areas. If several fields alone are examined a very misleading interpretation may result.

iii) Nasal tissue

It was only possible to carry out observations on the tissue eosinophilia in the allergic cases as they alone afforded sufficient biopsy material for both histamine determinations and pathological sections.

The sections were prepared by the Surgical Pathology Department of McGill University. They were stained with hematoxylin and eosin. (See Figure 5, Page 37).

The interpretation of the tissue eosinophilia is on a similar basis as that of the nasal smears. Here again we must consider the

general cellular picture of the stroma and report the degree of eosinophilia rather than the percentage (i.e. 0, +, ++ and +++). We must consider the whole section as the eosinophils again tend to cluster and may be very scanty or even absent in areas (Figures 6, 7, 8 and 9, Pages 39, 40, 41 and 42).

The blood smears, nasal smears and nasal tissue sections were all prepared from material taken between 8 a.m. and 11 a.m. This was done so that the normal diurnal variation of the blood eosinophils would be at a constant phase.

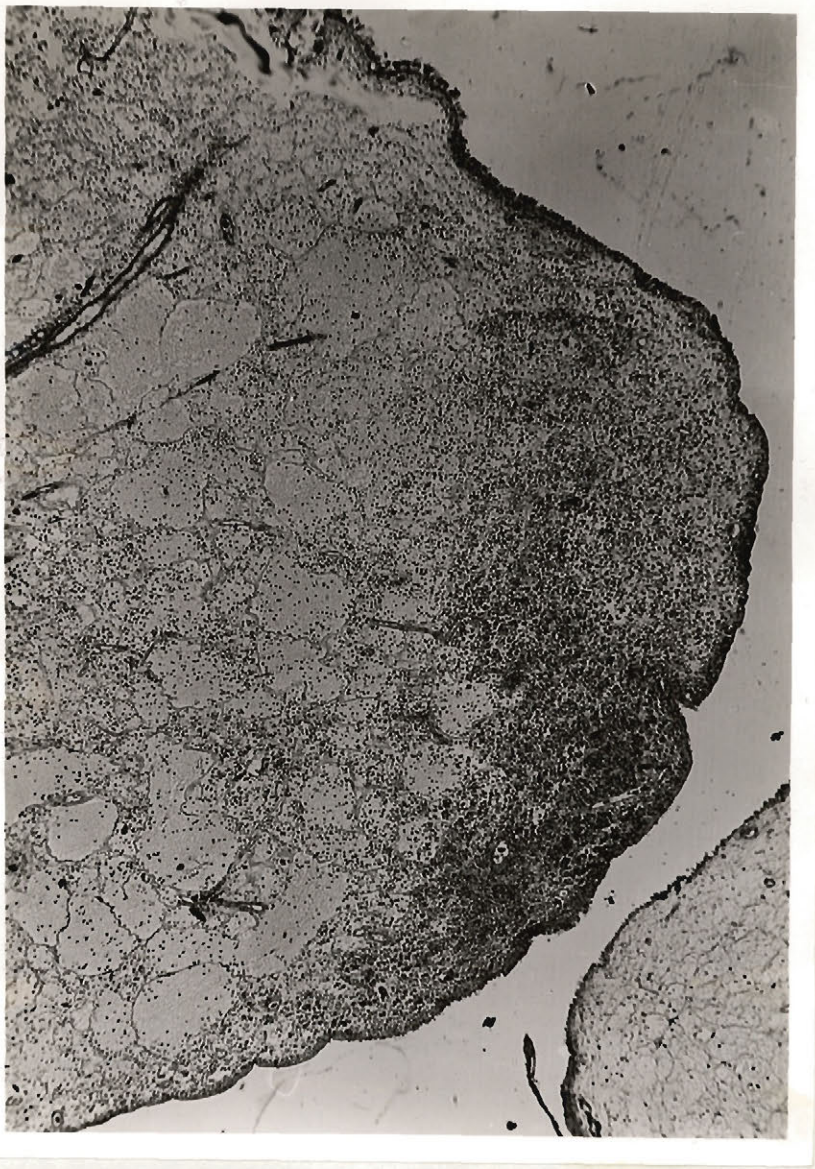


FIGURE 6

Nasal mucous polyp (A.L. - Surgical Pathology No. 51-5398 Polyp Histamine = 14.0 Y/gram). The stroma is heavily infiltrated with eosinophils. The variation in the degree of the cellular content in different areas is noticeable. (Magnification x 35).

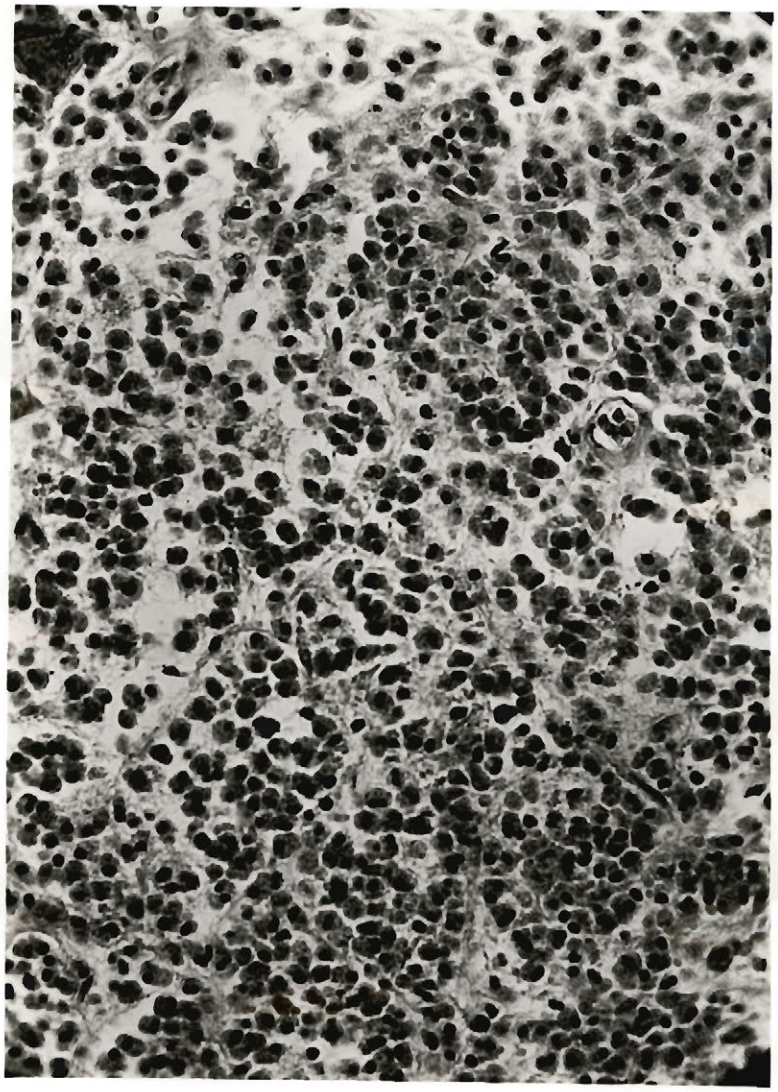


FIGURE 7

High magnification (x 370) of stroma of nasal mucous polyp (Figure 6 - Page 39). The majority of the cells are eosinophils. There are many mononuclear eosinophils present (see text - Page 30).

FIGURE 8

Nasal mucous polyp (H.S. - Surgical Pathology No. 51-5785 - Polyp Histamine = 11.78 γ/gram). The stroma has a uniform round cell content. (magnification x 100)

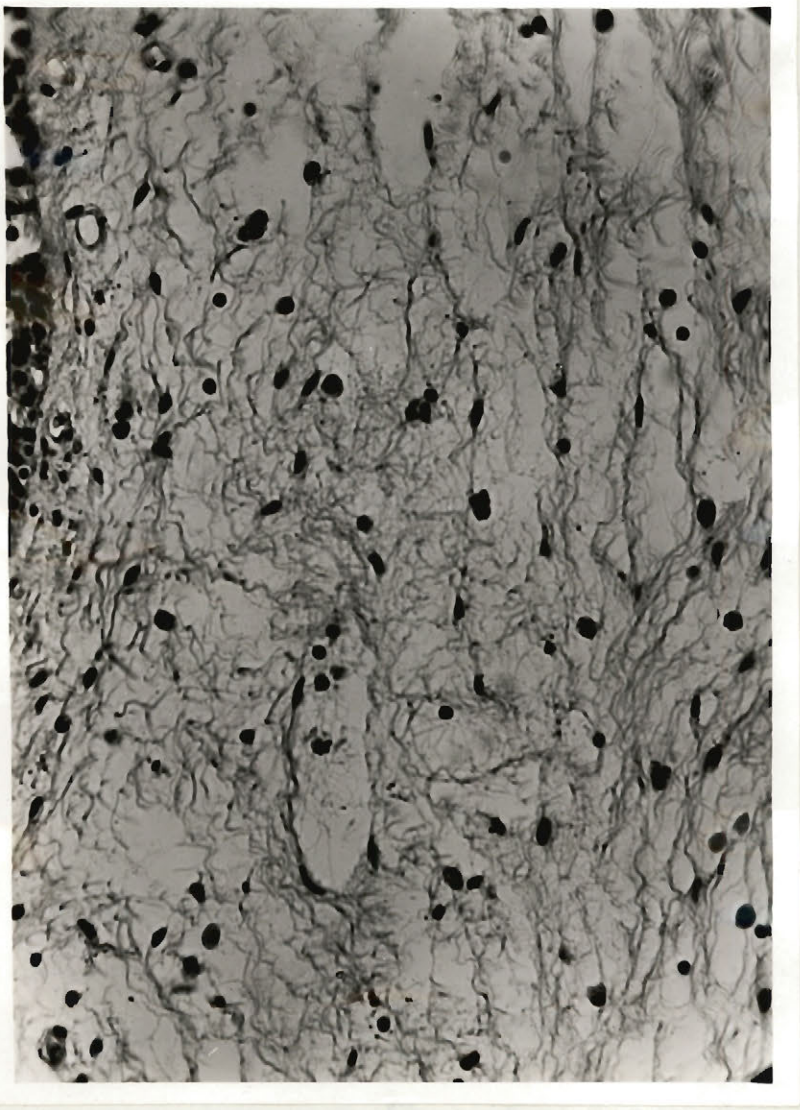


FIGURE 9

High magnification (x 370) of stroma of nasal mucous polyp (Figure 8 - Page 41) showing round cell content. No eosinophils are present.

NORMAL HUMAN NASAL MUCOUS MEMBRANE

A. Source, Selection and Division of Cases

The "normal" patients were all inpatients of the Department of Otolaryngology, Royal Victoria Hospital. They were cases either undergoing tonsillectomy, adenoidectomy, tonsillectomy and adenoidectomy, or submucous resection (designated in charts as T, A, T & A, and S.M.R. respectively). Except for the latter operation they were all subjected to general anaesthesia.

At the time of operation these patients were all asymptomatic as far as their local condition was concerned and all cases selected were free from any obvious complicating nasal conditions. We may consider them as normal individuals undergoing an elective operation.

The patients were carefully screened for any personal or family history of allergy. Blood and nasal smears were examined for eosinophils and if there was the slightest indication that any "allergic" tendencies existed they were excluded from the normal series.

In order to see if there were any variations in the histamine content of nasal mucous membrane at different age periods and with sex, the patients were divided into groups according to age and sex.

It has been shown by Rose (1939) that in normal rats the histamine content of various tissues is greater in males than in females, while that of the blood is greater in females than in males.

The age groups were as follows:

Group i) From birth to fourteen years of age - the child or pre-puberty group.

Group ii) From fifteen years to fourty-four years of age - the adult group.

Group iii) From fourty-five years of age and onwards - the elderly adult or post-menopausal group.

The ages of fourteen years and fourty-four years were selected as the dividing points of the groups because they are the average age for puberty and the menopause to occur respectively. Around these ages there are adjustments occurring in the endocrine system of the body and, associated with this, changes in tissue histamine values could conceivably take place.

B. Method of Obtaining Normal Nasal Mucous Membrane

The method of obtaining the normal nasal mucous membrane was identical in all cases.

At the termination of the scheduled operative procedure, while the patient was still under anaesthesia, mucous membrane biopsies were taken, using sterile precautions, from the middle third of the lower edge of the inferior turbinate. (See Figure 10, Page 45).

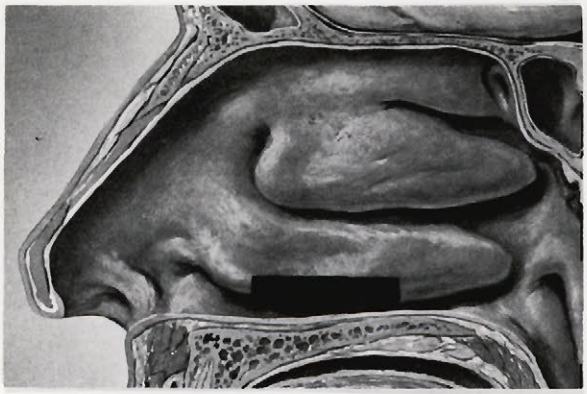
This was done under direct vision using a head mirror and reflexed light or an electric head light for illumination.

The anterior nares was dilated with an appropriate sized Vienna type nasal speculum and the inferior turbinate was visualized. The biopsies were taken with a small, angular, right or left handed Dabney nasal punch.

The amount of mucous membrane removed per punch was small (.003-.005 grams) and in order to obtain sufficient material for the histamine determinations it was necessary to remove several punches and pool them (resulting total .006-.015 grams).

In view of this I routinely removed a punch biopsy from each inferior turbinate of a patient.

Following this procedure there was usually a moderate degree of epistaxis but this was, in most cases, easily controlled and did not



Lateral view of nose showing shaded area on inferior turbinate from where biopsy material was removed.

FIGURE 10

necessitate nasal packing. This was especially true in the children. In several of the adults there was profuse nasal bleeding which necessitated packing of the nose and which prolonged their hospitalization.

The biopsy specimens were placed between layers of gauze moistened with normal saline. They were then transported as quickly as possible to the Medical Laboratory where the histamine determinations were performed.

C. Premedication and Anaesthetic Agents - Details and Discussion

The majority of the normal biopsies, including all those on the children, were taken while the patient was under general anaesthesia. In approximately one-third of the adults the procedure was performed under local anaesthesia.

The children were routinely premedicated with appropriate doses of atropine or morphine and atropine. The anaesthetic agents used were varying combinations of vinthene, nitrous oxide and ether, administered by intra-tracheal technique.

The adults undergoing general anaesthesia were premedicated with appropriate doses of morphine and atropine. They were induced by intravenous pentothal and curare and the anaesthesia maintained by intra-tracheal nitrous oxide, cyclopropane or trilene.

The adults undergoing local anaesthesia were premedicated with nembutal, morphine and hyoscine. The local anaesthesia was obtained by topical use of a 5% or 10% cocaine hydrochloride solution containing 15 drops to the half-ounce of Epinephrine 1:1000 solution.

We are markedly limited with patients in the degree to which we can pursue certain observations and as a consequence it was not possible to study the effect of anaesthesia as such on the histamine content of nasal mucous membrane. Nor could I find any references in the literature to suggest that there was any change in the histamine content of human nasal mucous membrane with various types of anaesthesia.

Vascular changes occur in the nasal mucous membrane during both general and local anaesthesia. They may be of a vasodilator type but the majority, and this is especially true of local anaesthesia with cocaine and epinephrine, are of a vasoconstrictor type.

If histamine was released from the cells under such circumstances you would expect some degree of vasodilatation to occur, in spite of any antagonistic effects of the premedication and anaesthetic agents.

Clinically, vasodilatation does not occur and, if anything, the tendency is towards a degree of vasoconstriction.

In view of this I feel there is no convincing clinical evidence to suggest that there is a release of histamine from the cells of the nasal mucous membrane during uncomplicated anaesthesia.

We will now consider the various groups of normal nasal mucous membrane.

D. Group I - the child or pre-puberty group. From birth to fourteen years of age.

i) Males Thirty individuals were observed ranging from two to twelve years of age. The following data was compiled:

TABLE I

Name	Age in years	Hospital no.	Operative Procedure	Blood Eosinophilia in %	Nasal smear Eosinophilia in degree	Histamine content Nasal Mucous Membrane in γ/gram
M.G.	4	51-14619	T & A	1	0	20.5
D.B.	7	51-14873	T & A	.5	0	10.4
P.L.	3	51-14876	T & A	1	0	27.8
K.D.	5	51-15259	T & A	2	0	13.9
G.M.	6	51-15261	T & A	1	0	15.0
B.C.	12	51-15633	T & A	1	0	7.5
G.B.	7	51-16328	T & A	0	0	54.0
S.F.	4	51-16782	T & A	1.5	0	28.6
C.N.	5	51-17082	T & A	.5	0	15.0
U.C.	4	51-17414	T & A	.5	0	20.8
M.C.	3	51-17415	T & A	0	0	12.1
L.B.	9	51-17503	T & A	-	-	20.8
W.B.	3	51-17679	T & A	1	0	22.8
C.W.	5	51-17681	T & A	3	0	26.3
M.K.	6	51-18135	T & A	1	0	17.3
D.P.	5	51-18212	T & A	.5	0	18.3
R.F.	4	51-18470	T & A	1.5	0	31.3
R.C.	6	51-18602	T & A	1	0	13.5
P.L.	7	51-18811	T & A	0	0	30.0
J.S.	5	51-18924	T & A	2	0	32.0
G.W.	8	51-19245	T & A	0	0	32.3
D.H.	4	52-151	T & A	1.5	0	15.0
P.F.	5	52-605	T & A	-	-	21.8
J.D.	7	52-819	T & A	0	0	17.6
A.L.	4	52-821	T & A	2	0	15.8
J.B.	2	52-1592	A	1	0	14.3
M.F.	4	52-1596	T & A	1	0	21.4
J.P.	8	52-2306	T & A	.5	0	22.7
M.T.	3	52-3390	T & A	1	0	5.0
A.A.	9	52-3402	T & A	1	0	13.8

The histamine determinations varied from 5.0 to 54.0 γ/gram of nasal mucous membrane. The average histamine value was 20.6 γ/gram.

The blood eosinophils were within normal values. The nasal smears were negative for eosinophils.

ii) Females Thirty individuals were observed ranging from three to twelve years of age. The following data was compiled:

TABLE II

Name	Age in years	Hospital no.	Operative Procedure	Blood Eosinophilia in %	Nasal smear Eosinophilia in degree	Histamine content Nasal Mucous Membrane in γ/gram
J.P.	9	51-17264	T & A	1	0	20.0
D.B.	6	51-17504	T & A	.5	0	19.7
D.R.	4	51-17552	T & A	2	0	13.3
J.L.	9	51-17822	T & A	1	0	11.1
P.L.	3	51-18025	T & A	1	0	15.0
C.D.	4	51-18086	T & A	2	0	17.3
H.F.	10	51-18471	T & A	0	0	16.7
J.S.	5	51-18536	T & A	2	0	9.4
L.C.	6	51-18645	T & A	.5	0	11.0
S.P.	6	51-18663	T & A	1	0	20.0
S.P.	8	51-18664	T & A	1	0	11.8
D.S.	5	51-19065	T & A	2	0	16.0
C.S.	6	51-19124	T & A	1	0	12.0
J.A.	3	51-19244	T & A	2	0	17.4
C.C.	5	51-19248	T & A	1	0	48.4
L.A.	6	51-19413	T & A	1.5	0	12.0
S.D.	4	51-19804	T & A	.5	0	31.2
I.D.	6	51-19809	T & A	2	0	41.7
L.C.	12	52-125	T & A	1	0	20.2
S.B.	4	52-382	T & A	2	0	14.2
B.M.	11	52-591	T & A	1	0	12.5
C.M.	7	52-592	T & A	-	-	30.0
A.S.	4	52-997	A	.5	0	17.5
C.P.	3	52-1381	T & A	1	0	15.0
C.H.	8	52-1590	T & A	2	0	27.7
H.M.	7	52-1926	T & A	1	0	6.0
J.H.	9	52-2311	T & A	2	0	16.2
L.L.	3	52-2388	T & A	-	-	26.5
L.P.	3	52-2392	T & A	1.5	0	27.5
M.R.	11	52-3187	T & A	3	0	6.3

The histamine determinations varied from 6.0 to 48.4 γ/gram of nasal mucous membrane. The average histamine value was 18.8 γ/gram.

The blood eosinophils were within normal values. The nasal smears were all negative for eosinophils.

iii) Discussion of Group I

The range of variation of the histamine content of normal nasal mucous membrane in this group for males (5.0 - 54.0 γ/gram) and females (6.0 - 48.0 γ/gram) is almost identical. On the average the males (20.6 γ/gram) have a slightly higher histamine content than the females (18.8 γ/gram). The average histamine content, regardless of sex, was 19.7 γ/gram.

In all cases the blood eosinophils were within normal values and the nasal smears were negative for eosinophils.

E. Group 2 - the adult group. From fifteen to fourty-four years of age.

i) Males Fifteen cases were observed ranging from eighteen to fourty-three years of age. The following data was compiled:

TABLE III

Name	Age in years	Hospital no.	Operative Procedure	Blood Eosinophils in %	Nasal smear Eosinophils in degree	Histamine content Nasal Mucous Membrane in γ/gram
M.C.	18	51-16271	S.M.R.	2	0	21.7
F.B.	33	51-17216	S.M.R.	1	0	8.3
V.Z.	26	51-17962	T & A	1.5	0	8.6
G.B.	32	51-18274	A	2	0	8.6
F.H.	18	51-18433	T & A	1	0	10.0
J.P.	43	51-18943	S.M.R.	1.5	0	20.5
W.K.	23	51-19745	T & A	2	0	11.2
P.C.	35	52-899	T & A	2	0	10.0
A.V.	28	52-1199	T & A	1	0	29.5
L.B.	35	52-1863	S.M.R.	1.5	0	20.1
F.L.	23	52-2052	S.M.R.	2	0	5.0
J.D.	38	52-2176	T & A	2	0	14.2
R.L.	32	52-2644	S.M.R.	3	0	6.25
T.G.	34	52-2789	S.M.R.	1	0	4.2
F.B.	37	52-3125	T & A	2	0	11.6

The histamine determinations varied from 4.2 to 29.5 γ/gram of nasal mucous membrane. The average histamine value was 12.7 γ/gram.

The blood eosinophils were within normal values. The nasal smears were all negative for eosinophils.

ii) Females Fifteen individuals were observed ranging from fifteen to thirty-nine years of age. The following data was compiled:

TABLE IV

Name	Age in years	Hospital no.	Operative Procedure	Blood Eosinophils in %	Nasal smear Eosinophils in degree	Histamine content Nasal Mucous Membrane in γ/gram
W.T.	36	51-14122	S.M.R.	3	0	19.6
G.N.	15	51-14812	T & A	0	0	8.2
S.C.	35	51-14816	T & A	1	0	26.5
E.V.	16	51-14942	S.M.R.	4	0	7.4
A.D.	21	51-15191	T & A	2	0	6.25
J.W.	16	51-17215	S.M.R.	2	0	33.4
M.U.	24	51-17629	T & A	1.5	0	27.8
D.D.	31	51-18150	T & A	1	0	14.0
J.M.	18	51-18290	T & A	2	0	16.5
J.D.	24	51-19485	T & A	1.5	0	17.0
E.S.	15	51-19739	T & A	1	0	28.4
G.V.	29	52-393	T & A	2	0	38.5
K.O.	39	52-1383	T & A	2	0	9.6
J.B.	25	52-1971	T & A	3	0	17.1
J.P.	16	52-2792	T & A	2	0	8.3

The histamine determinations varied from 6.25 to 38.5 γ/gram of nasal mucous membrane. The average histamine value was 18.6 γ/gram.

The blood eosinophils were within normal values. The nasal smears were all negative for eosinophils.

iii) Discussion of Group 2

The range of variation of the histamine content of normal nasal mucous membrane in this group for females (6.25 - 38.5 γ/gram) was

broader than that for the males (4.2 - 29.5 γ/gram). On the average the females (18.6 γ/gram) had a significantly higher histamine content than the males (12.7 γ/gram). The average histamine content, regardless of sex, was 15.7 γ/gram.

In all cases the blood eosinophils were within normal values. The nasal smears were negative for eosinophils.

F. Group 3 - the elderly adult or post-menopausal group. From fourty-five years of age and onwards.

This proved to be the most difficult group to obtain and only four cases were observed. These individuals were all in their fifties. Because of the small number of cases, no attempt was made to subdivide them according to sex. The following data was compiled:

TABLE V

Name	Sex	Age in yrs.	Hospital no.	Operative Procedure	Blood Eosinophils in %	Nasal smear Eosinophils in degree	Histamine content Nasal Mucous Membrane in γ/gram
J.M.	M	59	51-14141	Turbinectomy	2	0	1.2
E.T.	F	50	51-15263	S.M.R.	1.5	0	27.7
E.S.	M	57	51-18023	T & A	2	0	55.8
M.M.	F	50	51-18684	Antrum*	1	0	27.1

*Radical Antrum (Caldwell-Luc)

The histamine determinations in this group varied from 1.2 to 55.8 γ/gram of nasal mucous membrane. The average histamine value was 28.0 γ/gram.

The blood eosinophils were within normal values. The nasal smears were negative for eosinophils.

ii) Discussion of Group 3

From this data we can say that the histamine content of normal nasal mucous membrane in this group has a wide variation (1.2 to 55.8 γ/gram) and the indications are that the average is near the centre of this range (28.0 γ/gram).

In all cases the blood eosinophils were within normal values. The nasal smears were negative for eosinophils.

General Discussion - Normal Nasal Mucous Membrane

From these observations we can conclude that there is a marked variation in the histamine content of normal nasal mucous membrane.

These variations were:

Group I	5.0 - 54.0 γ/gram
Group 2	4.2 - 38.5 γ/gram
Group 3	1.2 - 55.8 γ/gram

The average histamine value for each group was:

Group I	19.7 γ/gram
Group 2	15.7 γ/gram
Group 3	28.0 γ/gram

According to sex the average histamine value for each group was:

	Males	Females
Group 1	20.6 γ/gram	18.8 γ/gram
Group 2	12.7 γ/gram	18.6 γ/gram
Group 3	-	-

In the males there is a decrease in the average histamine value in Group 2 as compared to Group 1, while in the females the values are almost identical.

In Group 1 the males have a slightly greater average histamine

value than the females while in Group 2 the females have an appreciably greater value than the males.

Two extremely high histamine values (83.3 and 198.4 γ/gram) were obtained during this study on males of Group 1. These values are not included in Table 1. Both of these cases had a completely negative personal and family history of allergy and were considered, at the time the biopsies were taken, "suitable" normals. The exact significance of these high values is not clear.

In all the normal cases the blood eosinophils were within normal values. The nasal smears were all negative for eosinophils.

In view of the fact that the majority, if not all nasal mucous polyps arise during adult life, it was decided to use the variation and average histamine content of Group 2 as the "Control Normals" (4.2 - 38.5 γ/gram and 15.7 γ/gram respectively).

In order to assess the role of certain factors on the histamine content of nasal mucous membrane the following series of cases were observed:

G. Negative Personal, Positive Family History of Allergy

i) Group 1 - Nine cases were observed ranging from three to eight years of age and the following data was compiled:

TABLE VI

Name	Sex	Age in years	Hospital no.	Operative Procedure	Blood Eosinophils in %	Nasal smear Eosinophils in degree	Histamine content Nasal Mucous Membrane in γ/gram
R.C.	M	7	51-17412	T & A	2	0	51.8
M.M.	F	4	51-18530	T & A	1.5	0	29.3
G.T.	F	8	52-2320	T & A	2	0	13.0
W.S.	F	4	52-2460	T & A	1	0	14.2
D.M.	M	3	52-3580	T & A	2	0	4.3
G.U.	M	3	52-3585	T & A	1.5	0	10.4
W.T.	M	5	52-3660	T & A	0	0	12.6
K.O.	F	4	52-3739	T & A	1	0	20.8
B.O.	M	4	52-3740	T & A	2	0	23.3

The histamine content of the nasal mucous membrane varied from 4.3 to 51.8 γ/gram. This is almost identical to the variation for Group 1 "normal" (5.0 - 54.0 γ/gram). The average histamine value was 19.9 γ/gram and this is again almost identical to the average value for Group 1 "normal" (19.7 γ/gram).

The blood eosinophils were within normal values and the nasal smears were negative for eosinophils.

ii) Group 2 - Three cases were observed ranging from nineteen to fourty-four years of age and the following data was compiled:

TABLE VII

Name	Sex	Age in years	Hospital no.	Operative Procedure	Blood Eosinophils in %	Nasal smear Eosinophils in degree	Histamine content Nasal Mucous Membrane in γ/gram
N.B.	F	22	52-1842	T & A	2	0	14.3
A.P.	M	49	52-2775	A	1	0	5.8
L.L.	F	19	52-2919	T & A	1.5	0	5.8

The histamine values were all within the "normal" range of variation for this group (4.2 - 38.5 γ/gram). The average histamine value was 8.6 γ/gram and this is below the average Group 2 "normal" (15.7 γ/gram). However, we must remember that this is only a small series.

The blood eosinophils were within normal values and the nasal smears were negative for eosinophils.

iii) Discussion

It appears, especially as shown in Group 1 of these observations, that there is no difference between the nasal mucous membrane histamine content of individuals with "allergic tendencies" and "normals".

H. Chronic Infection - Negative Personal or Family History of Allergy

i) Group 2 - Three cases were observed ranging from twenty-four to thirty-three years of age. The tissue examined was removed during radical antral surgery for chronic infection which had no allergic component. The following data was compiled:

TABLE VIII

Name	Sex	Age in years	Hospital no.	Operative Procedure	Blood Eosinophils in %	Nasal smear Eosinophils in degree	Histamine content Nasal Mucous Membrane in γ/gram
E.W.	F	27	51-19676	Antrum	2	0	37.5
B.J.	M	33	52-372	Antrum	1	0	24.1
O.G.	M	24	52-2371	Antrum	2	0	1.32

The histamine values were all within the normal range for this group (normal: 4.2 - 38.5 γ/gram). They averaged 20.9 γ/gram (normal: 15.7 γ/gram).

The blood eosinophils were within normal values and the nasal smears were negative for eosinophils.

ii) Discussion

From this small series it is difficult to draw any definite conclusions except that there is no marked change in the histamine content of the nasal mucosa in non-allergic patients with a chronic infection.

I. Chronic Infection - Positive Nasal Allergy

Two allergic cases (H.L. 51-16899 and H.S. 51-19076) with nasal mucous polyps and an associated antral infection were observed. These patients had radical sinus surgery performed and the histamine values of the antral mucosa were 40.4 and 9.3 γ/gram respectively. These values are similar to normal Group 2 (4.2 - 38.5 γ/gram) and indicate that such coexisting conditions do not lead to any marked change in the tissue histamine.

General Discussion Sections G,H and I

From these observations we can conclude that the histamine content of the nasal mucous membrane is not altered from the normal range of variation by inherited allergic tendencies, chronic infections or chronic infections associated with nasal allergy.

We do not know why extremely high histamine values are encountered on nasal mucous membrane in the occasional "normal" individual.

ALLERGIC HUMAN NASAL MUCOUS MEMBRANE - NASAL MUCOUS POLYPS

A. Source of Cases

It is considered today that nasal mucous polyps are all allergic in origin. In view of this and the relative ease of obtaining biopsy material from such cases this work was carried out on patients with nasal mucous polyps.

These patients, as already mentioned, were observed through either the Allergic Clinic or the Department of Otolaryngology, Royal Victoria Hospital.

As many cases as possible were studied and no attempt was made to divide them according to age or sex.

B. Method of Obtaining Nasal Mucous Polyps

The nasal mucous polyps were all obtained by essentially the same method. They were removed, under local anaesthetic, with a nasal snare.

This was done under direct vision using a head mirror and reflexed light or an electric head light for illumination.

The procedure, depending upon its extent, was performed on an out-patient or in-patient basis. It is usually a simple, uncomplicated matter to remove several small mucous polyps on an ambulatory patient but if any extensive removal is contemplated, it is safer to have the individual hospitalized. Occasionally the post-operative course can be complicated by nasal haemorrhage and under suitable circumstances this is easily controlled.

The tissue, following removal, was handled in the same manner as the normal nasal mucous membrane.

C. Premedication and Anaesthesia

The out-patients were not premedicated. The in-patients were premedicated with appropriate doses of nembutal, morphine and hyoscine or morphine and atropine.

Local anaesthesia was obtained by the topical use of a 5% or 10% solution of cocaine hydrochloride containing 15 drops to the half ounce of epinephrine 1:1000 solution.

In all cases a minimal amount of the anaesthetic agent was used and attempts were made to apply it directly to the stock of the polyp.

In order to study the effect of anaesthesia on the histamine content of polyps, in a patient with multiple polyps one was removed without anaesthesia and was immediately followed by further removal under anaesthesia.

No significant change was found to occur in the polyp histamine under these varied conditions.

D. Nasal Mucous Polyps

In all, 56 cases with nasal mucous polyps were studied (35 males, 21 females) and 59 separate operative procedures were performed.

The following data was compiled:

CHART I

NAME	SEX	AGE IN YEARS	HOSPITAL NO.	SURGICAL PATHOLOGY NO.	PERSONAL HISTORY OF ALLERGY	FAMILY HISTORY ALLERGY	DURATION OF NASAL SYMPTOMS	DEGREE OF NASAL SYMPTOMS	DEGREE OF NASAL SIGNS	BLOOD EOS. %	NASAL SMEAR EOS.	TISSUE EOS.	HISTAMINE CONTENT OF POLYPS γ/Gm
S.H.	F	68	A6504	51-4112	Yes	No	10 years	mild	mild	1	++	+++	30.4
M.G.	F	35	51-14417	51-4236	Yes	No	7 years	mild	mild	1	0	+	12.6
M.G.	F	35	B5052	51-5061	Yes	No	7 years	mod.	mod.	4	++	++	9.1
E.W.	M	37	51-14420	51-4235	Yes	Yes	14 years	mild	nil	1	0	0	23.1
*F.S.	F	67	G9693	51-4252	Yes	Yes	25 years	mild	mild	3	+	+++	22.0
*F.S.	F	67	G9693	51-4647	Yes	Yes	25 years	severe	severe	6	+	+++	6.2
*F.S.	F	67	G9693	-	Yes	Yes	25 years	severe	severe	6	++	+++	5.8
F.B.	F	70	C5887	-	Yes	No	1 year	nil	nil	1	0	-	14.0
A.B.	F	60	B6330	51-4406	Yes	No	45 years	mild	mild	0	+	++	12.5
A.B.	F	60	B6330	51-4733	Yes	No	45 years	mild	mild	9	+	+	17.5
K.G.	F	55	J42	-	Yes	No	12 years	mod.	mod.	7	+	-	16.1
A.S.	M	28	51-15331	51-4494	Yes	No	8 years	severe	mod.	3	0	+	6.67
E.B.	F	15	51-15373	51-4528	Yes	Yes	1 year	nil	nil	2	+	+	41.5
A.A.	M	48	J2538	51-4565	Yes	No	40 years	nil	nil	3	++	+++	13.7
L.F.	M	46	H344	-	Yes	-	10 years	nil	nil	1	0	-	14.2
J.M.	M	29	OPD-Priv.	51-4799	Yes	-	6 years	nil	nil	2	+	++	14.7
M.H.	M	54	OPD-Priv.	51-4874	Yes	No	30 years	mild	mild	1	0	0	10.5

*Discussed in text

CHART II

NAME	SEX	AGE IN YEARS	HOSPITAL NO.	SURGICAL PATHOLOGY NO.	PERSONAL HISTORY ALLERGY	FAMILY HISTORY ALLERGY	DURATION OF NASAL SYMPTOMS	DEGREE NASAL SYMPTOMS	DEGREE NASAL SIGNS	BLOOD EOS. %	NASAL SMEAR EOS.	TISSUE EOS.	HISTAMINE CONTENT OF POLYPS γ/Gm.
J.H.	M	63	C7803	-	Yes	No	31 years	nil	nil	2	+	-	13.8
A.G.	M	53	51-16670	51-4914	Yes	-	25 years	mod.	mod.	1	+	+	9.6
B.F.	M	39	51-16898	51-4967	Yes	No	6 years	mild	mild	5	++	+	17.8
H.L.	M	42	51-16899	51-4969	Yes	No	30 years	severe	severe	8	+	+	10.2
W.R.	M	50	51-17229	51-5174	Yes	No	22 years	nil	nil	0	-	0	24.8
A.S.	M	69	51-17432	51-5106	Yes	No	6 months	mod.	mod.	1	-	+	7.3
U.B.	F	57	51-17510	51-5233	Yes	Yes	10 years	mod.	mod.	4	0	+	6.8
G.W.	M	45	51-17539	51-5148	Yes	Yes	15 years	mild	mild	3	++	+	15.3
L.S.	F	55	C8124	51-5175	Yes	Yes	1 year	severe	severe	2	0	++	3.8
Y.V.	F	38	J6771	51-5287	Yes	No	8 years	mild	mild	6	+	+	20.0
*S.R.	F	46	J5831	51-5371	Yes	Yes	2 years	severe	severe	2	+++	+++	7.8
*S.R.	F	46	J5831	-	Yes	Yes	2 years	mild	mild	3	+++	-	22.2
A.L.	F	61	51-18153	51-5398	Yes	Yes	9 years	mild	mild	-	-	+++	14.0
A.T.	M	41	51-18268	51-5505	not proven	No	8 years	mild	mild	1	+	+	14.7
J.C.	M	27	51-18273	51-5521	Yes	No	11 years	mild	mild	1	0	+	16.7
E.S.	F	39	51-18355	51-5496	Yes	Yes	7 years	severe	severe	.5	+	++	7.7
C.V.	M	19	K2002	51-5506	Yes	Yes	4 years	mild	mild	1	+	+	22.7

*Discussed in text

CHART III

NAME	SEX	AGE IN YEARS	HOSPITAL NO.	SURGICAL PATHOLOGY NO.	PERSONAL HISTORY ALLERGY	FAMILY HISTORY ALLERGY	DURATION OF NASAL SYMPTOMS	DEGREE NASAL SYMPTOMS	DEGREE NASAL SIGNS	BLOOD EOS. %	NASAL SMEAR EOS.	TISSUE EOS.	HISTAMINE CONTENT OF POLYPS Y/Gm.
A.S.	M	48	G6060	51-5523	Yes	Yes	12 years	nil	nil	3	+	+	15.8
H.S.	M	48	51-19076	51-5785	Yes	No	1 year	mod.	mod.	2	+++	+	11.7
A.L.	M	41	51-19258	51-5766	Yes	Yes	8 years	nil	nil	2	+	+++	21.9
S.T.	M	17	51-19746	51-5906	Yes	Yes	2 years	mod.	mod.	20	+	++	16.7
D.D.	M	68	OPD-Priv.	51-5916	Yes	Yes	40 years	mod.	mod.	-	-	++	11.6
M.C.	F	52	H8139	52-37	Yes	Yes	14 years	severe	severe	2	++	+++	7.4
M.P.	M	24	52-53	52-99	Yes	Yes	3 months	mild	mild	3	+	+++	48.9
L.P.	M	48	52-402	52-102	Yes	No	1 year	mild	mild	1	0	++	15.9
J.L.	M	34	52-664	52-170	Yes	Yes	10 years	mod.	mod.	1	+	+	11.4
R.D.	M	54	OPD-Priv.	52-331	Yes	Yes	10 years	severe	severe	2	++	+++	2.63
R.C.	M	42	52-1055	52-533	Yes	Yes	6 months	mild	mild	11	+	+	12.0
A.W.	M	72	52-1726	52-547	Yes	Yes	30 years	mod.	mod.	6	+	+	9.26
P.Z.	M	28	52-1929	52-620	Yes	No	3 years	mod.	mod.	1	0	+	20.9
E.Y.	F	43	52-2219	52-791	Yes	Yes	12 years	nil	nil	2	0	0	12.7
D.J.	F	18	52-2527	-	Yes	Yes	14 years	mod.	mod.	2	+	-	21.8
R.T.	M	50	OPD-Priv.	52-876	Yes	-	6 months	mod.	mod.	2	0	0	6.0
L.H.	M	30	OPD/Priv.	52-893	Yes	No	1 year	mod.	mod.	1	++	+	4.2
S.S.	M	67	52-2794	52-908	Yes	Yes	15 years	mild	nil	1	0	+	9.0

CHART IV

NAME	SEX	AGE IN YEARS	HOSPITAL NO.	SURGICAL PATHOLOGY NUMBER	PERSONAL HISTORY ALLERGY	FAMILY HISTORY ALLERGY	DURATION OF NASAL SYMPTOMS	DEGREE NASAL SYMPTOMS	DEGREE NASAL SIGNS	BLOOD EOS. %	NASAL SMEAR EOS.	TISSUE EOS.	HISTAMINE CONTENT OF POLYPS γ/Gm.
R.W.	M	53	52-2991	52-936	Yes	No	1 year	mild	mild	2	+	+++	24.8
J.W.	M	49	52-3059	52-965	Yes	No	27 years	mod.	mod.	2	+	+	8.38
J.P.	M	57	52-3177	52-998	Yes	No	17 years	severe	severe	3	+	+	7.8
B.G.	F	50	52-3212	52-1020	Yes	No	17 years	severe	severe	4	+	++	3.1
P.P.	F	36	52-3331	52-1067	Yes	No	1 year	mild	mild	1	0	+	17.5
P.B.	M	50	B8272	52-1049	Yes	-	15 years	severe	severe	3	+	+	2.6
*T.K.	F	40	51-16201	51-4773	Yes	-	3 years	mod.	mod.	-	-	+	108.0

*Not included in general series but discussed in text.

The cases ranged from fifteen to seventy years of age. Except for one case they all had definite proven allergy. The one exception, although no definite allergy could be proven, was strongly suggestive of such an etiology. Half of the cases had a positive family history while the other half had a negative family history of allergy. They were all cases of perennial allergy with nasal symptoms varying from 6 months to fourty-five years.

The histamine content of the polyps varied from 2.6 to 48.9 γ/gram with an average of 14.3 γ/gram. This range is broader than Group 2 normal (4.2 - 38.5 γ/gram) but the average values are quite similar (Group 2 normal: 15.7 γ/gram).

From these figures it appears that there is no significant difference by wet weight of tissue in the histamine content of nasal mucous polyps when compared as such with that of normal nasal mucous membrane. This confirms the work of Buhrmester and Wenner (1936). However, in such comparisons no provision was made for the degree of the allergic reaction present at the time the tissue was removed.

The degree of the allergic reaction in the nasal mucosa of patients with nasal allergy, with or without mucous polyps, is variable. At periods, depending upon several factors, the allergic reaction may be at a minimum or even quiescent, while at other times it is quite severe.

The degree of nasal signs and symptoms varies directly with the degree of the allergic reaction occurring in the nose. If the allergic reaction is severe the nasal signs and symptoms are severe and vice versa. Critical observations of the degree of nasal signs and symptoms can therefore be used as an indication of the degree of the allergic reaction occurring in the nose.

This is essentially true in cases with nasal mucous polyps but here we have the added factor of a mechanical blockage of the nasal passages by the polypoidal masses. In certain cases, although the allergic reaction may be quiescent, the patients complain of marked nasal blockage which is purely on a mechanical basis. In assessing nasal polyp cases, one must remember this point and pay particular attention to the condition of the mucous membrane as such.

In this study the degree of nasal signs and symptoms present at the time the nasal polyps were removed was recorded and efforts were made to remove tissue during various phases of the disease process.

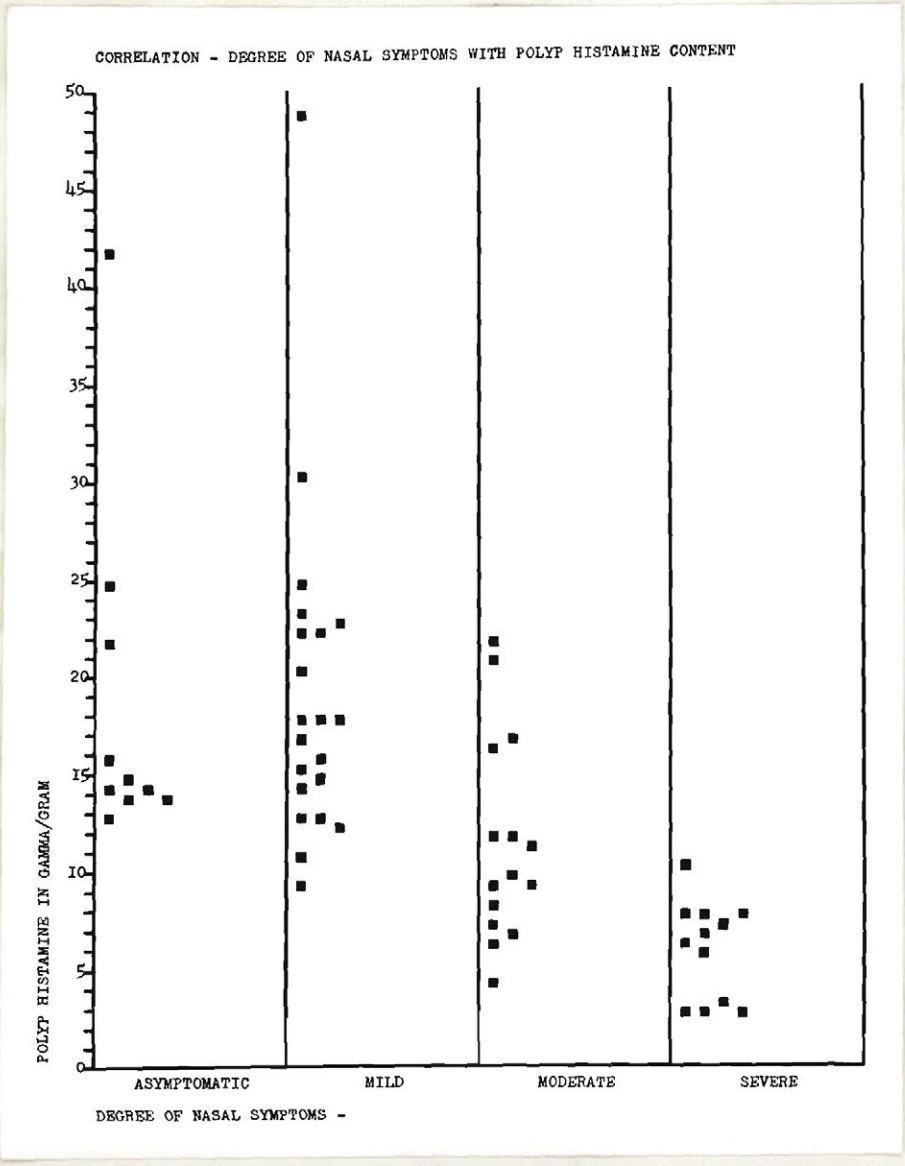
The degree of nasal signs and symptoms ("degree of allergic reaction"), was reported as asymptomatic or nil, mild, moderate or severe.

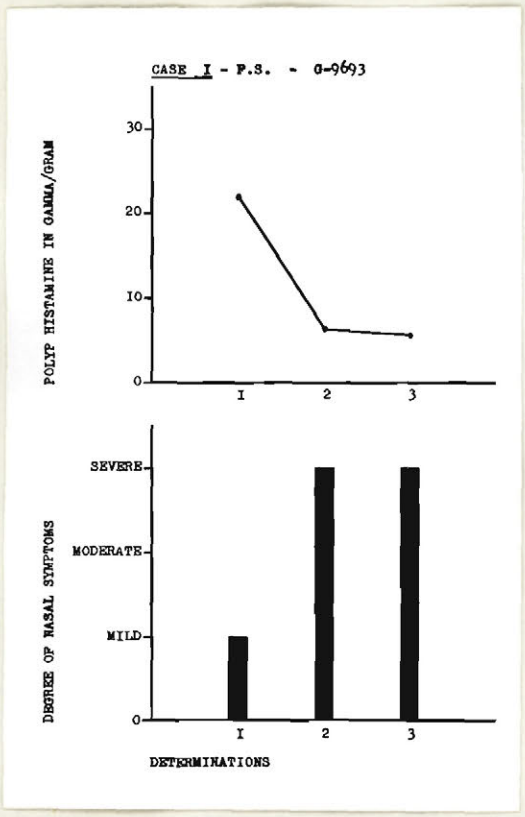
The chart shown on Page 67, correlating the degree of nasal signs and symptoms ("Symptoms" in charts) with the polyp histamine content, was prepared from the data.

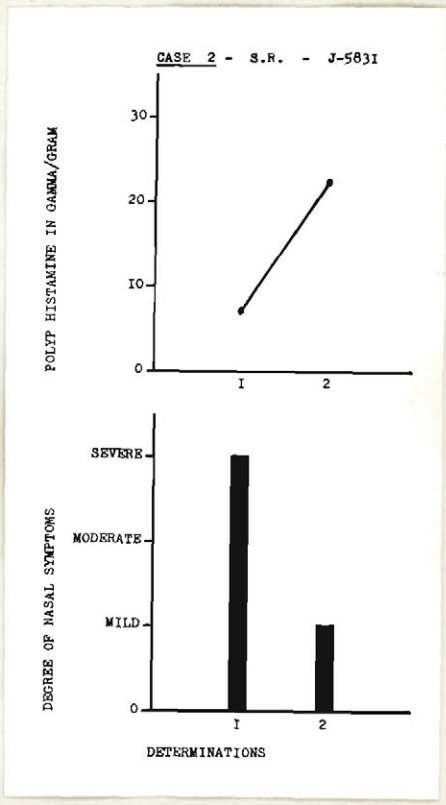
It is obvious from this chart that with increasing degrees of nasal signs and symptoms there is a decrease in the histamine content of the nasal polyps. That is, with an increase in the severity of the allergic reactions the histamine content of the polyps decreases.

The opportunity arose with two patients to obtain biopsy material during different phases of their nasal signs and symptoms. The charts shown on Pages 68 were prepared.

CASE I - Three different observations were made - one during mild symptoms and two during severe symptoms. With the severe symptoms there was a marked decrease in the histamine content of the mucous polyps (From 22.0 to 6.2 and 5.8 γ/gram).


CORRELATION - DEGREE OF NASAL SYMPTOMS WITH POLYP HISTAMINE CONTENT
POLYP HISTAMINE IN GAMMA/GRAM
0
5
10
15
20
25
30
35
40
45
50
ASYMPTOMATIC
MILD
MODERATE
SEVERE
DEGREE OF NASAL SYMPTOMS -





CASE I - F.S. -9693 CASE II - S.R. J-5831

CASE II - Two observations were made - one during severe symptoms and one during mild symptoms. The histamine content of the tissue was low during the severe symptoms (7.8 Y/gram) and was high during the mild symptoms (22.2 Y/gram).

Discussion Polyp Histamine

From these observations one concludes that there is a decrease in the histamine content of nasal mucous polyps with increasing severity of the allergic reaction.

Although there is a certain dilution factor involved in such a decrease, this is not a major factor as shown by simultaneous studies on wet weight and dry weight histamine determinations on nasal mucous polyps.

This finding tends to collaborate the cellular theory of the allergic mechanism which postulates, during the reaction, the release of preformed histamine from the cells of the shock organ through stimulation or injury by the antigen-antibody complex. If histamine was released from the cells under such circumstances you would expect to find, for a certain period, a decrease in the cellular histamine.

Katz (1942) demonstrated in patients showing a skin reaction to the intradermal injection of ragweed, that there was an increase in the output of histamine from the skin into which the antigen had been injected. This increased output of histamine rose sharply within a few minutes after injection of the antigen and fell to the original level within sixty minutes.

Rose (1941) showed with patients suffering from dermographia and cold allergy that during the formation of extensive wheals histamine may be liberated from the skin and appear for brief periods in increased quantities in the blood.

These observations demonstrate a release of histamine from the cells into the interstitial fluid and blood and its eventual diffusion away from the shock organ.

During these studies no observations were made on the actual shock organ "tissue" but it is conceivable that under such circumstances one would find a decrease in the cellular histamine content of such tissue.

Pellerat and Murat (1946) demonstrated that a decrease in the histamine content of skin occurs after freezing and that the local diminution in histamine results from its release into the blood.

Such a decrease could be expected to occur in allergic situations where the shock organ was exposed to repeated and prolonged "antigen insults".

In all the mucous polyps studied during this work the allergic reaction had been present, in varying degrees, for an extended period. No case was observed during the initial onset of an allergic state nor during the reactivation of a quiescent allergic nasal condition.

This work demonstrates a decrease in the histamine content of nasal mucous polyps with increasing severity of the allergic reaction over a prolonged period.

During this study one allergic case with moderate nasal signs and symptoms was observed who had a polyp histamine value of 108.0 γ/gram. As already mentioned, two normal male children were observed with nasal mucous membrane histamine values of 83.3 and 198.4 γ/gram. Rose, Entin and Baxter (1951), in their small series on nasal tissues, came across two such high values on antral mucous membrane from allergic individuals (101 and 140 γ/gram).

It was thought initially that such high values may be associated with pre-existing allergic tendencies, chronic infection or chronic infection associated with allergy, but this has been shown during the investigation not to be the case. The exact significance of such high histamine values on nasal mucous membrane in both normal and allergic individuals is not clear.

Eosinophils

In the normal cases no blood or nasal smear eosinophilia was encountered. Insufficient biopsy material was obtained for simultaneous observation on the tissue eosinophils.

In the allergic cases alterations on the eosinophils were noted. Blood eosinophilia existed in many cases and varying degrees of nasal smear and tissue eosinophilia was found to be the rule in the majority of the cases.

From these observations it seems that the demonstration of eosinophils in nasal smears is good presumptive evidence for the existence of nasal allergy. As no observations were carried out during this study on normal nasal tissues no conclusions should be drawn on the significance of the tissue eosinophilia observed in the majority of the allergic patients. However, it has been known for years (Bizzozero (1887), Gollasch (1889), Seiffert and Kahn (1895), Huber and Koessler (1922), Hansel (1929-30), Cameron (1935) and Eggston and Wolff (1947)) that allergic reactions are associated with local infiltration of eosinophils and normally they do not exist in any quantity in tissues. So it appears safe to say that the tissue eosinophilia observed is also good presumptive evidence of allergy.

Cowie and Jimenez (1936) found, in a study on almost one hundred cases, that simultaneous differential counts on blood smears and nasal smears did not show any definite relationship to each other.

From my data there is also no correlation between simultaneous observations on blood, nasal smear or tissue eosinophilia.

There is also no correlation between the degree of tissue eosinophilia and the histamine content of the nasal mucous polyps.

Hansel (1933-34, 1936) infers that there is a direct relationship between the degree of nasal symptoms and the degree of nasal smear eosinophilia.

From the data the following charts have been prepared:

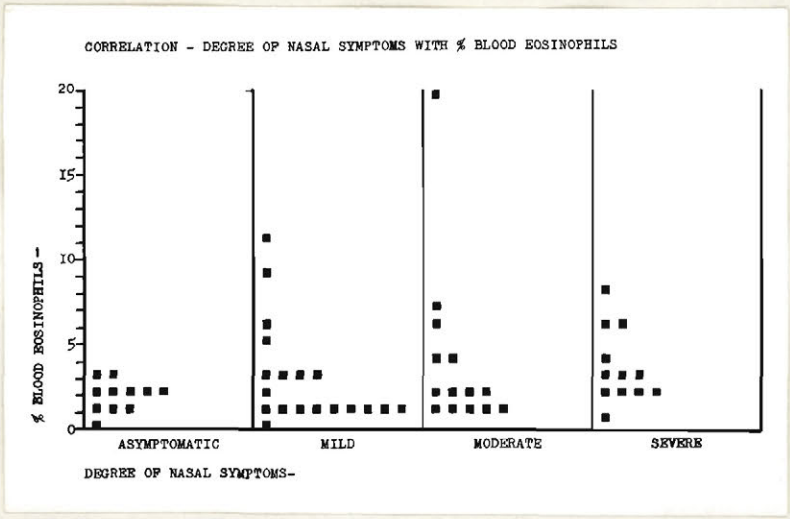


CHART I

Correlation of the Degree of Nasal Symptoms with the % of Blood Eosinophils.

There is no correlation between the degree of nasal symptoms and the % of blood eosinophils.

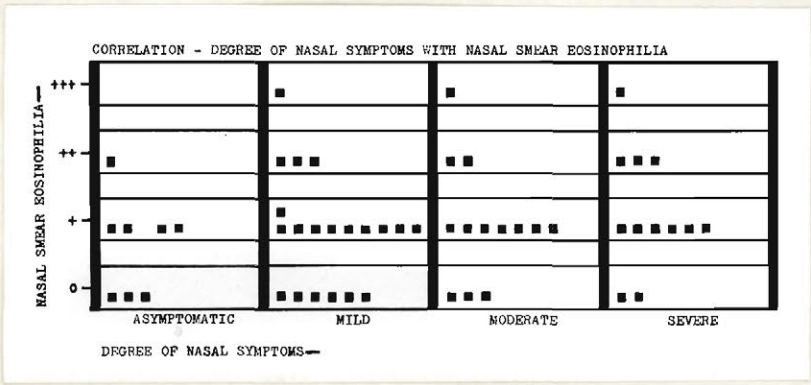


CHART II

Correlation - Degree of Nasal Symptoms with the Degree of Nasal Smear Eosinophilia.

There is no correlation between the degree of nasal symptoms and the degree of nasal smear eosinophilia.

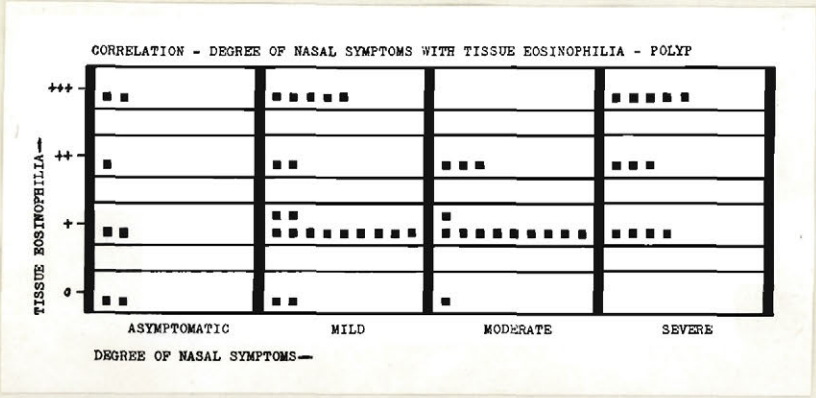


CHART III

Correlation - Degree of Nasal Symptoms with the Degree of Tissue Eosinophilia

There is no correlation between the degree of nasal symptoms and the degree of tissue eosinophilia.

Discussion - Eosinophils

The demonstration of eosinophils in nasal smears and tissue is good presumptive evidence for the existence of nasal allergy. However, it should not be considered a definite diagnostic indicator of allergy as they may be present in conditions other than allergic.

There is no correlation between simultaneous observations on blood, nasal smear or nasal tissue eosinophilia or between the degree of nasal signs and symptoms and the blood, nasal smear or nasal tissue eosinophilia.

There is no correlation between the histamine content of the mucous polyps and the tissue eosinophilia.

Summary

This clinical investigative work has lead to the following findings:-

i) There is a wide variation in the histamine content of normal nasal mucous membrane. There are slight differences in this variation according to age and sex groups but the overall normal variation is from approximately 5 to 50 ϒ/gram of histamine by wet weight of tissue.

ii) In normal children the males have a slightly higher average nasal mucous membrane histamine value than the females while in the adults the females have a significantly higher average histamine value than the males. The average female histamine value for children and adults is approximately the same.

iii) The nasal mucous membrane of individuals with allergic tendencies, chronic infections and chronic infections associated with nasal allergy have histamine values within the normal range of variation.

iv) The variation of the histamine content of nasal mucous polyps (allergic nasal mucous membrane) on the basis of wet weight is similar to that of the normal nasal mucous membrane and the average histamine values are almost identical.

v) On the basis of the degree of the allergic reaction there is a definite decrease in the histamine content of nasal mucous polyps with a severe reaction.

vi) In both normal nasal mucous membrane and nasal mucous polyps an occasional high histamine value is encountered, the exact significance of which is not clear.

vii) The finding of eosinophils in nasal smears and nasal tissue

is good presumptive evidence for the existence of nasal allergy but it is not a definite diagnostic criteria.

viii) There is no correlation between simultaneous observations on blood, nasal smear and tissue eosinophilia in cases of nasal allergy.

ix) There is no correlation between the degree of nasal allergy and the blood, nasal smear and tissue eosinophilia.

x) There is no correlation bwtween the degree of tissue eosinophilia and the histamine content of the nasal mucous polyps.

BIBLIOGRAPHY

Abel, J.J. and Kubota, S. On the presence of histamine in tissue. Jour. Pharm., 13; 243; 1919.

Abell, R.G. and Schenck, H.P. Microscopic observations on the behaviour of living blood vessels of the rabbit during the reaction of anaphylaxis. Journ. Immunol., 34; 195; 1938.

Adams, H.M., Hunter, R.B. and Kinnear, T.W.S. Observations on the urinary excretion of histamine in urticaria. Quart. J. Exp. Physiol., 36; 49; 1950.

Airila, V. Uber die Ursache der Todlichen Blutdrucksenkung im Akuten Anaphylaktischen Shock beim Kaninchen. Skandinav. Arch. f. Physiol., 31; 388; 1914.

Anderson, J.F. and Rosenau, M.J. Anaphylaxis. Arch. Int. Med., 3; 519; 1909.

Answers to Queries: Eosinophilia. J.A.M.A., p.1034; Dec. 1949.

Auer, John and Lewis, P.A. The physiology of the immediate reaction of anaphylaxis in the guinea pig. Jour. Exper. Med., 12; 151; 1910.

Auer, J. Lethal cardiac anaphylaxis in the rabbit. Fourth communication. Jour. Exper. Med., 14; 476; 1911.

Barger, G. and Dale, H.H. B-iminazolethylamine and the other active principles of ergot. Proc. Chem. Soc., 26; 128; 1910.

Barger, G. and Dale, H.H. B-iminazolethylamine, a depressor constituent of intestinal mucosa. Jour. Phys., 41; 499; 1911.

Bartosch, R., Feldberg, W. and Nagel, E. Das Freiwerden eines Histaminahnlichen Stoffes bei der Anaphylaxie des Meerschweinchens. Arch. f. d. ges. Physiol., 230; 129; 1932.

Bartosch, R. Uber die Herkunft des Histamins bei der Anaphylaxie des Meerschweinchens. Klin. Wchnschr., 14; 307; 1935.

Best, C.H., Dale, H.H., Dudley, H.W. and Thorpe, W.V. The nature of the vaso-dilator constituents of certain tissue extracts. Jour. Phys., 62; 397; 1927.

Best, C.H. and McHenry, E.W. Inactivation of histamine: method of histamine estimation. Jour. Phys., 70; 349; 1930.

Bezancon, F. and Bernard, E. L'Eosinophilie dans l'asthme, ses equivalents et ses parentesmorbides. Ann. de Med., XXVII; 204-33, 1930.

Biedl, A. and Kraus, R. Experimentelle Studien uber Anaphylaxie. Wien. Klin. Wchnschr., 22; 363; 1909.

Billroth, C.A. General Surgical Pathology. Appleton, New York, 1883.

Bizzozero, G. Handbuch der Klin. Mikroscopie, Erlangen, 1887.

Buhrmester, C.C. and Wenner, W.F. Presence of Histamine-like substance in nasal mucosa, nasal polypi and nasal secretions. Arch. Otolaryng., 24; 570-581; 1936.

Cameron, J.A.M. An investigation of the part played by allergy or sensitization as a factor in predisposing the mucous membrane of the nasal passages, and the paranasal sinuses to infection and its bearing upon the treatment of diseases of these cavities. Jour. Laryng. and Otol., 50; 493-511; 1935.

Cloetta, M. and Anderes, E. Besitzen die Lungen Vasomotoren? Arch. f. Exper. Path. u. Pharmakol., 76; 125; 1914.

Coates, G.M. and Ersner, M.S. Occurrence of eosinophiles in the mucous membrane of the maxillary sinus in asthmatic patients. Arch. Otl., 11; 158; 1930.

Coca, A.F. The site of reaction in anaphylactic shock. Ztschr. f. Immunitatsforsch., 20; 622; 1914.

Coca, A.F. The mechanism of the anaphylaxis reaction in the rabbit. Jour. Immunol., 4; 219; 1919.

Code, C.F. and Ing, H.R. The isolation of histamine from the white cell layer of centrifuged rabbit blood. Jour. Physiol., 90; 501; 1937.

Code, C.F. The histamine content of the blood of guinea pigs and dogs during anaphylactic shock. Am. Jour. Physiol., 127; 78; 1939.

Cowie, D.M. and Jimenez, B. Cytologic examination of nasal smears of sensitized and nonsensitized persons with nasal symptoms with special reference to the eosinophil count and to simultaneous blood counts. Arch. Int. Med., 57; 75; 1936.

Dale, H.H. and Laidlaw, P.P. The physiological action of B-iminazolethylamine. Jour. Physiol., 41; 318; 1910.

Dale, H.H. The anaphylactic reaction of plain muscle in the guinea pig. Jour. Pharmacol. and Exp. Therap., 4; 167; 1913.

Dale, H.H. and Dudley, H.W. Presence of histamine and acetylcholine in spleen of ox and horse. Jour. Phys., 68; 97; 1929.

Daly, I. de B. and Schild, H.O. Inactivation by histaminase preparations of the histamine-like substance recovered from lungs during anaphylactic shock. J. Physiol., 83; 3P, 1934.

Daly, I. de B., Peat, S. and Schild, H.O. Release of histamine-like substance from lungs of guinea-pigs during anaphylactic shock. Quart. J. Exptl. Physiol., 25; 33; 1935.

Denecke, Gerhard. Ueber die Bedeutung der Leber fur die Anaphylaktische Reaktion beim Hunde. Ztschr. f. Immunitatsforsch. u. Exp. Therap., 20; 501; 1914.

Deschamps, J.L. Traite des maladies des fosses nasales. Richards, Paris, 1804.

Dragstedt, C.A. and Gebauer-Fuelnegg, Erich. Studies in Anaphylaxis: I. The appearance of a physiologically active substance during anaphylactic shock. Am. Jour. Physiol., 102; 512; 1932.

Dragstedt, C.A. and Mead, F.B. Farther observations on the nature of the active substance ("Anaphylatoxin") in canine anaphylactic shock. Jour. Immunol., 30; 319; 1936.

Dragstedt, C.A. and Mead, F.B. The role of histamine in canine anaphylactic shock. Jour. Pharmacol. and Exp. Therap., 57; 419; 1936.

Dragstedt, C.A. and Mead, F.B. Peptone shock. Jour. Pharmacol. and Exp. Therap., 59; 429; 1937.

Dragstedt, C.A., Ramirez de Arellano, Max, and Lawton, A.H. The Relationship of histamine to anaphylaxis in the rabbit. Sci. n.s., 91; 617; 1940.

Dragstedt, C.A., Ramirez de Arellano, Max, Lawton, A.H. and Youmans, G.P. Passive sensitization of rabbits' blood. Jour. Immunol., 39; 537; 1940.

Drinker, C.K. and Bronfenbrenner, J. The pulmonary circulation in anaphylactic shock. Jour. Immunol., 9; 387; 1924.

Eggston, A.A. i)Pathology of accessory sinus disease. Trans. Amer. Laryng. Assoc., 52; 181; 1930.

Eggston, A.A. ii) Pathology of chronic sinusitis. Arch. Otol., 12; 561; 1930.

Eggston, A.A. iii) Maxillary sinusitis. Pathology., New York State J. Med., 33; 508; 1933.

Eggston, A.A. and Wolff, D. Histopathology of the ear, nose and throat. Williams and Wilkins, Baltimore, 1947.

Ehrlich, P. Verhandlung des Physiologischen Gesellschaft zu Berlin, 1878-79.

Ewing, J. Neopastic diseases, 1940, 4th edition. Saunders, Philad. and London.

Eyermann, C.H. Nasal manifestations of allergy. Ann. Otol., Rhinol. and Laryngol., 36; 808; 1927.

Fabricant, N.D. Relation of pH of nasal secretions in situ to activity of lysozyme; report of case of experimentally induced allergic rhinitis. Arch. of Otl., 41; 53-55; 1945.

Feldberg, W. Histamine and anaphylaxis. Ann. Rev. Physiol., 3; 671; 1941.

Finck, H.P. Tissue changes in the nasal mucosa. Laryngoscope, 37; 783; 1927.

Gaddum, J.H. and Dale, H.H. Gefasserweiternde Stoff. der Gewebe, Leipzig, 1936.

Gaddum, J.H. The metabolism of histamine. B.M.J., Oct. 27, 1951.

Gay, F.P. and Southard, E.E. Further studies in anaphylaxis: iv) the localization of cell and tissue anaphylaxis in the guinea pig, with observations on the cause of death in serum intoxication. Jour. Med. Research, 19; 17; 1908.

Gebauer-Fuelnegg, Erich and Dragstedt, C.A. Studies in anaphylaxis: ii) the nature of a physiologically active subsfance appearing during anaphylactic shock. Am. Jour. Physiol., 102; 520; 1932.

Gollasch: Zur Kenntnis der Asthmatischen Sputums, Fortschr. d. Med., 7; 361; 1889.

Gottesman, J.M. and Gottesman, J. Use of Histamine as standard test for diminished resistance in suprarenalectomized rats. J. Exper. Med., 47; 503; 1928.

Gotzl, F.R. and Dragstedt, C.A. Peptone shock in rabbits. Jour. Pharmacol. and Exp. Therap., 74; 33; 1942.

Guggenheim, M. and Loffler, W. Biochem. Ztschr., 72; 303; 1916.

Hanke, M.T. and Koessler, K.K. Studies on proteinogenous amines; quantitative colorimetric estimation of histamine in protein and protein-containing matter. Jour. Biol. Chem., 43; 543; 1920.

Hansel, F.K. Clinical and histopathologic studies of the nose and sinuses in allergy. J. Allergy, 1; 43-70; 1929-30.

Hansel, F.K. Observations on the cytology of secretions in allergy of the nose and paranasal sinuses. Jour. Allergy, 5; 357; 1933-4.

Hansel, F.K. Allergy of the nose and paranasal sinuses. St. Louis, C.V. Mosby Co., 1936.

Hansel, F.K. Cytologic observations on secretions of nose and paranasal sinuses in allergy. Tr. Am. Laryng. Assoc., 62; 293-339; 1940.

Hansel, F.K. Allergy in otolaryngology. Laryngoscope, 58; 652-672; 1948.

Hawkins, D.F., Mongas, J.L. and Schild, H.O. Histamine release from asthmatic human lung. J. Physiol., 113; 26P, 1951.

Haworth, E. and MacDonald, A.D. On histamine in cotton dust, and in the blood of cotton workers. Jour. Hyg., 37; 234; 1937.

Heineke, A. and Deutschmann, F. Munch. Med. Wschr., LIII: 797-8; 1906.

Hirsch, O. Nasal polypi - their origin and its solution. Bull. Battle Creek Sanitar. and Hosp. Clin., 24; 7; 1929.

Hoppmann, C.M. Ueber Nasenpolypen. Monatschr. f. Ohrenh., 19; 161; 1885.

Huber, H.L. and Koessler, K.K. The pathology of bronchial asthma. Arch. Intern. Med., 30; 689-760; 1922.

Jaques, L.B. and Waters, E.T. The identity and origin of the anti-coagulant of anaphylactic shock in the dog. Jour. Physiol., 99; 454; 1941.

Johnson, M.C. and Goldstein, D.W. Allergy and the cytologic examination of nasal smears. Arch. Otl., 16; 808; 1932.

Jones, Warthin. Philosophical Trans., London, p.63; 1846.

Kahn, I.S. and Stout, B.F. Cytologic examination of nasal smears in the differential diagnosis of allergy and infection. South. Med. Jour., 25; 583; 1932.

Katz, Gerhard. Histamine release from blood cells in anaphylaxis in vitro. Sci., N.S., 91; 221; 1940.

Katz, G. and Cohen, S. Experimental evidence for histamine release in allergy. J. Amer. Med. Ass., 117; 1782; 1941.

Katz, Gerhard. Histamine release in allergic skin reaction. Proc. Soc. Exp. Biol. and Med., 49; 272; 1942.

Kern, R.A. and Schenck, H.P. Allergy constant factor in etiology of so-called mucous polyps. J. Allergy, 4; 485-497; 1932-33.

Kern, R.A. and Schenck, H.P. Importance of allergy in etiology and treatment of mucous polyps. J.A.M.A., 103; 1293-1298; 1934.

Kern, R.A. and Schenck, H.P. Diagnosis and treatment of mucous polyps, with consideration of allergic factor. M. Clin. North America, 22; 1633-1643; 1938.

Lewis, Thomas and Grant, R.T. Vascular reactions of the skin to injury: ii) the liberation of a histamine-like substance in injured skin; the underlying course of factitious urticaria and of wheals produced by burning; and observations upon the nervous control of certain skin reactions. Heart, 11; 209; 1924.

Lewis, T. The blood vessels of the human skin. (London 1927).

Mann, A. and Lehmann, H. The eosinophil level in psychiatric conditions. Canad. M.A.J., 66; 52; 1952.

Manwaring, W.H. Serophysiologische Untersuchungen: i) Der Physiologische Mechanismus des Anaphylaktischen Shocks. Ztschr. f. Immunitatsforsch. u. Exp. Therap., 8; 1; 1910.

Manwaring, W.H., Hosepian, V.M., O'Neill, F.I. and Moy, H.B. Hepatic reactions in anaphylaxis: x) the hepatic anaphylatoxin. Jour. Immunol., 10; 575; 1925.

Morrison, W.W. Disease of the ear, nose and throat. New York, Appleton-Century-Crofts, Inc., 1948.

Mullin, W.V. and Ball, R.P. Studies of pathologic tissue removed from chronically infected nasal accessory sinuses. Ann. Otol., Rhinol. and Laryngol., 37; 192; 1928.

Ojers, Gaylord, Holmes, C.A. and Dragstedt, C.A. The relation of the liver histamine to anaphylactic shock in dogs. Jour. Pharmacol. and Exp. Therap., 73; 33; 1941.

Opie, E.L. Amer. J. Med. Sci., CXXVII; 217-39; 1904.

Otto, R. Quoted by Anderson, J.F. and Rosenau, M.J. Anaphylaxis. Arch. Int. Med., 3; 519; 1909.

Pellerat, J. and Murat, M. Histamine content of the skin: its variation under the influence of cold, during tuberculin allergy and in certain dermatoses. Ann. de Dermat. et Syph., 6; 76-84; 1946.

Polson, C. Contribution to discussion on nasal allergy (excluding hay-fever). J. Laryng. and Otol., 58; 224; 1943.

Rawlins, A.G. Chronic allergic sinusitis. Laryngoscope, 57; 381-399; 1947.

Richet, Charles-Robert. Anaphylaxis. (Translated by J.M. Blight) Liverpool, University Press, 1913.

Roche e Silva, M. Anaphylaxis in the rabbit. Jour. Immunol., 38; 333; 1940.

Rose, B. and Browne, J.S.L. Distribution and rate of disappearance of intravenously injected histamine in rats. Am. Jour. Physiol., 124; 412; 1938.

Rose, B. Metabolism of histamine - Ph. D. thesis. McGill University, page 135-135A; 1939.

Rose, Bram and Weil, Paul. Blood histamine in rabbit during anaphylactic shock. Proc. Soc. Exp. Biol. and Med., 42; 494; 1939.

Rose, Bram. The relationship of histamine to anaphylaxis and allergy. McGill Med. Jour., 10; 5; 1940.

Rose, Bram. Blood and tissue histamine during rabbit anaphylaxis. Am. Jour. Physiol., 129; 450; 1940.

Rose, Bram. Studies on blood histamine in cases of allergy: ii) Alteration in the blood histamine in patients with allergic disease. Jour. Clin. Inves., 20; 419; 1941.

Rose, Bram. Studies on blood histamine in cases of allergy: i) Blood Histamine during wheal formation. Jour. Allergy; 12; 327; 1941.

Rose, Bram, Rusted, I. and Fownes, J.A. Intravascular catheterization studies of bronchial asthma. i) histamine levels in arterial and mixed venous blood of asthmatic patients before and during induced attacks. Jour. Clin. Inves., 29; 1113-1119; 1950.

Rose, Bram, Pare, J.A.P., Pump, K.K., Stanford, R.L. and Johnson, L.G. Influence of ACTH on the excretion of histamine and histidine in patients with allergic states or rheumatoid arthritis. J. Clin. Inves., 29; 841; 1950.

Rose, Bram, Entin, M.M. and Baxter, H. (In press) as quoted by Rose, B: Allergy, Ann. Rev. Med., 2; 155; 1951.

Schild, H.O. Release of histamine-like substance in anaphylactic shock from various organs of the guinea-pig. (Abstr.) Jour. Physiol., 90; 34P; 1937.

Schild, H.O. Histamine release in anaphylactic shock from various tissues of the guinea-pig. Jour. Physiol., 95; 393; 1939.

Schmidt, A.D. Zur Kenntnis der Asthma Bronchiale. Zentralbl. f. Inn. Med., 12; 473; 1891.

Schultz, W.H. Physiological studies in anaphylaxis: i) the reaction of smooth muscle of the guinea-pig sensitized with horse serum. Jour. Pharmacol. and Exp. Therap., 1; 549; 1910.

Seiffert, O. and Kahn, M. Atlas der Histopathologie der Nase. Wiesbaden, J.F. Bergmann, 1895.

Serafini, U.J. Studies on histamine and histamine antagonists. J. Allergy, 19; 256; 1948.

Sewall, E.C. and Hunnicott, M.D. Cytologic examination of the antram. Arch. Otl., 10; 1; 1929.

Shambaugh, G.E., Jr. Nasal allergy for the practising rhinologist. Annals of Otol., Rhinol. and Laryngol., 54; 43; 1945.

Sherman, J.L. The eosinophil as an aid in clinical diagnosis. U.S. Armed Forces Med. Jour., 2; 383; 1951.

Thorpe, W.V. Isolation of histamine from heart. Biochem. J., 24; 626; 1930.

Ungar, G. and Parrot, J.L. Rechercher sur le choc anaphylactique in vitro. Mise en liberte d'une substance active par le poumon isole du cobaye sensibilise. Compt. rend. soc. biol., 123; 676; 1936.

Unger, L. Bronchial asthma. Charles C. Thomas, Publisher, 1945.

Virchow, Archiv. f. Path. Anat., 93; 213; 1883.

Voegtlin, Carl and Bernheim, B.M. The liver in its relation to anaphylactic shock. Jour. Pharmacol. and Exp. Therap., 2; 507; 1911.

Von Pirquet, C. Allergie. Berlin, Julius Springer, 1908.

Wachstein, M. Des Freiwerden eines Histaminahnlichen Stoffes bei der Anaphylaxie des Meerschweinchens. Arch. f. d. ges. Physiol. (Pflugers), 231; 24; 1932.

Walsh, T.E. and Lindsay, J. i) Cytology of nasal polyps, Arch. Otl., 20; 649; 1934.

ii) Nasal secretions: the value of cytologic examination to the rhinologist. Arch. Otl., 17; 783; 1933.

Watanabe, K. Der Gehalt der Hundeorgane an Darmkontrahierenden und Blutdrucksenkenden Stoffen in Stadium der Eiweiss-Sensibilisierung und im Anaphylaktischen Shock. Ztschr. f. Immunitatsforsch. u. Exp. Therap., 73; 149; 1931.

Weichselbaum, A. Grundriss der Pathologischen. Histolgie, Leipzig, 1892.

Weil, Richard. The vasomotor depression in canine anaphylaxis. Jour. Immunol., 2; 429; 1917.

Weil, Richard. Studies in anaphylaxis: xxi) Anaphylaxis in dogs. A study of the liver in shock and in peptone poisoning. Jour. Immunol., 2; 525; 1917.

Weil, Richard and Eggleston, Cary. Studies in anaphylaxis: xxii) Anaphylactic reactions of the isolated dogs' liver. Jour. Immunol., 2; 571; 1917.

Weille, F.L. Asthma: xi) The pathology of allergic tissue as seen in the nose and in the accessory sinuses. Arch. Otl., 12; 785; 1930.

Windaus, A. and Vogt, W. Ber. d. Duet. Chem. Gesellschaft., 4; 636; 1907.

Wright, J. The so-called hyaline bodies and other cellular degenerations in nasal polyps. Am. Jour. Med. Sci., 116; 445; 1898.

Wymon, L.C. Studies on suprarenal insufficiency: Anaphylaxis in suprarenalectomized rats. Amer. Jour. Phys., 89; 356; 1929.

Yonge, E.S. The determining cause of nasal polyps. B.M.J., 2; 964; 1907.

Zinsser, H., Enders, J.F. and Fathergill, D. Immunity, Principles and application in medicine and public health. 5th ed., the MacMillan Co., New York., 1939.