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THE EFFECT OF X-IRRADIATION ON TISSUE HISTAMINE IN THE RAT

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In the present work we have studied the effects of irradiation on the histamine content of the skin and alimentary canal in rats, and on the histological appearance of these tissues, particularly of their mast cells. Since both histamine and heparin are constituents of the mast cells a few experiments were also made on the blood coagulation time before and after irradiation.

Irradiation produces characteristic sequences of changes in the tissues of all animals. Changes in the histological appearance of the tissues have been widely investigated and attempts have been made to correlate these changes with the features of the radiation syndrome (Bloom, 1947). Metabolic changes following irradiation have also been investigated, and attempts have been made to correlate them with the clinical features (Prosser, 1947).

There have been few investigations on the effect of irradiation on the histamine content of the tissues, although it has been suggested that histamine liberated from irradiated cells or possibly newly formed from irradiated histidine in the tissues may be responsible for some of the effects of radiation (Ellinger 1951). Weber & Steggerda (1949) have shown that there is an increase in blood histamine values following irradiation in the rat, and Venters & Painter (1950) state that dogs and rabbits become more sensitive to histamine infusion following irradiation. Feldberg & Loeser (1954) have, in two human cases, demonstrated that the skin histamine is reduced following irradiation, and Bryant, Eisen, Ellis & Wilson (1955) have also shown changes in the histamine content of the skin and of the wall of the upper part of the alimentary canal in the rat after total body irradiation.

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METHODS

Radiation techniques and doses

Male albino rats weighing about 200 g were used. Before and after radiation they received rat cake and water *ad lib.* The animals which received total body radiation had no special treatment. Those which received local radiation of the abdomen were anaesthetized with pentobarbitone (Nembutal, Abbott Laboratories,) 40 mg/kg by intraperitoneal injection. The abdomens of the rats in which local skin reactions were observed were shaved before radiation: chemical depilators were not used. Any rats with visible damage to the skin after shaving were discarded. The animals were replaced in their cages and they were randomly selected at intervals after irradiation for investigation. They were investigated in groups of three at a time and so all observations were made on at least three rats and generally on six or nine rats.

The radiation conditions in the experiments were as follows: 200 kV, 10 mA, half-value layer 1·3 mm of copper, 40 cm focal skin distance. The exposure rate was 29 r/min, estimated at the surface of the skin nearest to the applicator.

Rats which were given total body irradiation received 1025 r on the skin of the back and 605 r on the abdominal skin. Each rat occupied a stall in a partitioned Perspex box with walls 5 mm thick, the ends of which were drilled for ventilation. The box was placed at the end of a 15 by 20 cm applicator.

Rats which received local radiation of the abdomen were irradiated whilst wearing a 3 mm lead shield covering pelvis and chest and head with an aperture of 5 × 4·5 cm over the abdomen. They received a dose of 1000 r to the abdominal skin. For observation of local reactions of skin vessels anaesthetized rats were irradiated singly with their shaved abdomens in contact with the end of a 3 cm diameter circular applicator of focal skin distance 27 cm. These rats received a dose of 2000 r to the abdominal skin; treatment time was 34 min.

Estimation of tissue histamine

The skin. Immediately after death the abdominal skin was shaved and a sample of skin weighing about 500 mg was taken from the left side of the middle line of the abdomen. The skin sample was split from the underlying tissue on the surface of the muscle along the natural line o cleavage, weighed and immediately boiled in 2 ml./g of N-HCl. It was then ground with sand and subsequently treated by the method of Feldberg & Talesnik (1953), except that the suspension was centrifuged and the supernatant fluid was decanted and diluted appropriately with Tyrode solution.

The stomach and jejunum. Immediately after removal of the skin the abdomen was opened and the stomach and jejunum removed. The posterior wall of the portion of the stomach lying between the pylorus and fundus, known as the pyloric part of the stomach, was removed. It was spread out, washed gently with a stream of saline, dried on filter-paper and about 500 mg were weighed and immediately boiled in N-HCl, 2 ml./g. The tissue was then extracted for histamine in the same way as the skin. The mesentery was dissected from the jejunum. The first two cm were used for histological examination. The rest of the jejunum was split open, washed gently with saline and then dried in filter-paper. About 500 mg of the proximal portion were weighed and immediately boiled in N-HCl and treated like the skin and stomach.

The extracts were assayed on the atropinized guinea-pig ileum and the results expressed in μg histamine base/g tissue. The usual control experiments were carried out to exclude from the assay substances other than histamine which cause contraction of the gut. In the jejunum very low values were obtained following irradiation, and part of the contraction was due to such substances which it was not possible to eliminate by dilution.

Blood coagulation time

The coagulation time was measured in groups of three rats before irradiation and at various periods after irradiation. Blood was obtained from the tail vein after the tail had been held in

warm water for 2–3 minutes. The coagulation time was estimated by the method of Griffith & Farris (1942). The end-point of coagulation is difficult to define accurately and consequently the error of its measurement tends to be high (Cohn, 1952).

Histological techniques

To demonstrate mast cells in subcutaneous tissue spreads, skin was removed from the right side of the abdomen near the mid-line and its subcutaneous tissue removed from the deep surface, mounted as described by Riley (1953) and the spreads were then stained with 0·5% toluidine blue. Paraffin sections were made from the anterior wall of the pyloric portion of the stomach and the first 2 cm of jejunum. One half of each tissue was fixed in formol saline and stained with haematoxylin and eosin, the other half was fixed in absolute alcohol and stained with toluidine blue.

Local reactions of skin vessels

Twenty hours after irradiation with 2000 r, pontamine sky blue was injected into the tail vein and immediately afterwards 0·1 ml. of saline, histamine, or compound 48/80 was injected intradermally into the centre of the irradiated area and also into normal skin over the lower part of the thorax. Histamine was injected in concentrations of 1/1000, 1/20,000, 1/30,000 and 1/50,000, and compound 48–80 in concentrations of 1/2000, 1/10,000 and 1/50,000. Fifteen minutes later the animal was killed and the skin was dissected off the abdomen and chest and mounted on a glass slide and frozen. The extent and intensity of the blueing in the normal and irradiated skin could then be compared. It was found that killing the animal 15 min after injection of the drugs produced optimal blueing with the concentrations used. The interval between the intradermal injections into the normal and irradiated skin was less than 30 sec, and the order of injections did not influence the results.

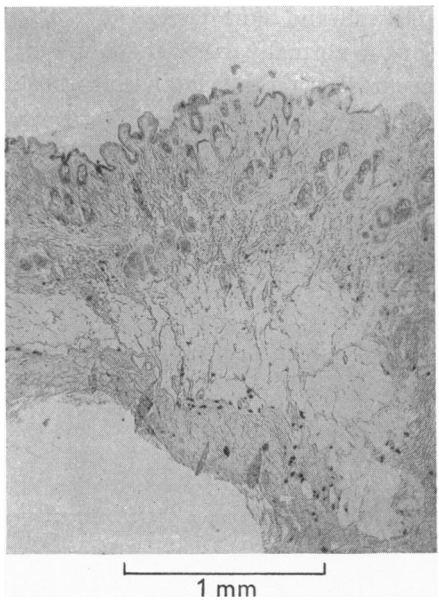
RESULTS

The effect of total body irradiation

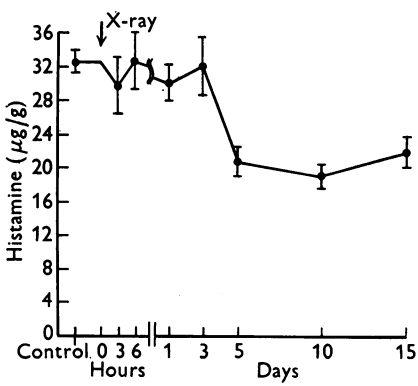
The dose of 1025 r total body irradiation applied to twelve rats caused the death of three of them after 15 days and of five in 25 days. The animals lost weight during the week following irradiation, but in the animals which survived this weight loss was subsequently regained.

Histamine content of the skin. Riley & West (1953) have shown that the histamine content of various tissues, including the skin, is closely related to the numbers of mast cells which they contain. Depending therefore on the thickness of the skin, and the quantity of subcutaneous tissue which is removed with it, the histamine content of the skin will vary. In the present experiments, as far as possible, skin of the same thickness and skin from the same part of the abdomen was always used. In Text-fig. 1 there is shown a section in order to demonstrate the thickness of the skin which was used and its content of mast cells. The mean value for the histamine content of the abdominal skin from eleven rats was 32·6 μg/g with a standard error of 1·3. The effect of 605 r on the histamine content of the abdominal skin is shown in Text-fig. 2. Throughout the first 24 hr and on the third day after the irradiation the histamine value remained within the normal range, but on the fifth post-irradiation day it fell, and reached the minimum value of 18·9 μg/g on the tenth day. Thereafter it increased slightly. The observations at each time period were made on at least six rats.

Histamine content of the stomach and jejunum. The histamine content of the pyloric portion of the stomach in eight rats was 23·4 μg/g with a standard error of 1·7. It began to decrease during the first 24 hr after total body irradiation with 1025 r and reached its minimum value on the fifth day (Text-fig. 3). Thereafter it increased slightly but it was still significantly below the control value 15 days after irradiation. The observations at each time period were made on three rats.



Text-fig. 1. Section of normal abdominal skin showing the thickness which was used for histamine estimations and the mast cell content. Toluidine blue.



Text-fig. 2. Effect of 605 r X-irradiation on the histamine content of abdominal skin. The standard error of the observations at each time interval is shown.

The mean value for the histamine content of the jejunum obtained from the control rats was 19·0 μg/g with a standard error of 1·5. Three hours after irradiation the histamine content had already begun to diminish, by the end of 24 hr it had fallen to 5·5 μg/g, and on the fifth post-irradiation day it had fallen almost to its minimum value. On the tenth day the histamine content had fallen to 1·4 μg/g at which it remained (Text-fig. 3).

The changes in the histamine contents of the different tissues following total body irradiation have been compared in Text-fig. 4. Whereas the histamine content of the stomach wall and skin decreased to about 60%, that of the jejunum fell to 7·5% of the normal value. The figure also illustrates that the changes in the skin occur considerably later than those in the stomach and jejunum.

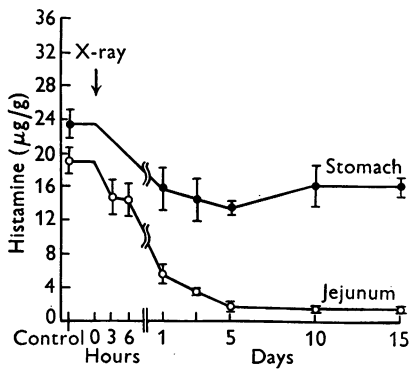


Fig. 3

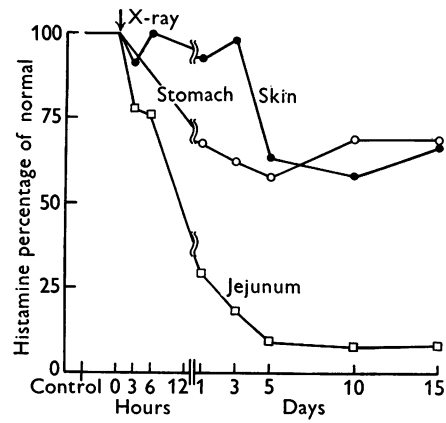


Fig. 4

Text-fig. 3. Effect of 1025 r total body X-irradiation applied to the skin of the back on the histamine content of the stomach and jejunum. The standard error of the observations at each time interval is shown.

Text-fig. 4. The changes in the histamine content of the abdominal skin, the stomach, and the jejunum of the rat following 1025 r total body X-irradiation, expressed as percentages of the normal values.

The epithelium of the stomach and jejunum. The cells in the wall of the stomach passed through the sequence of changes which normally follow irradiation (Pierce, 1948). At the end of 24 hr the mucous cells forming the gastric pits and the necks of the mucous glands were poorly stained and the nuclei were pyknotic. The granules in the zymogenic cells were pale and amorphous and in most of the parietal cells there was vacuolization of the cytoplasm and the nuclei had disappeared or were pyknotic. Mitoses appeared in the cells during the following days, and by the fifth post-irradiation day the mucous neck cells were reappearing and there were many fresh cells in the bases of the gastric glands. Numerous mitoses were present in the zymogenic

cells and their granules had reappeared. The parietal cells had resumed their normal appearance.

Twenty-four hours after irradiation there was degeneration of the columnar cells in the crypts of the jejunum, particularly in their superficial portions. There was pallor of the cytoplasm and many nuclei showed pyknosis. The goblet cells showed vacuolization. The lamina propria was oedematous and many of the cells were shrunken. On the third and fifth post-irradiation days there was progressive growth of the deeply staining columnar cells from the bottom of the crypts. The goblet cells at the bases of the glands were stained normally and contained granules. The columnar epithelium at the surface was degenerating and sloughing off into the lumen of the canal. On the tenth day deeply staining columnar cells extended more than half the distance from the bases of the glands to the surface, and there were numerous goblet cells amongst them. Cells of Paneth had reappeared at the bases of the glands. The lamina propria was still slightly oedematous and pyknotic nuclei were visible in it but it was beginning to resume its normal appearance.

The mast cells of the subcutaneous tissue. The changes in the appearance of the mast cells after irradiation resembled those described by Riley & West (1955*a*) following the injection of 'subacute doses' of compound 48/80 intraperitoneally into rats, except that they occurred more gradually. Twenty-four hours after irradiation the majority of the cells showed normal staining reactions and contained tightly packed granules. A few of the cells were swollen and the granules were less tightly packed than in normal cells. On the fifth post-irradiation day (Pl. 1, fig. 1*a*) the mast cells were swollen and the cell membranes of many of them were ruptured. Granules had leaked out of the cells into the surrounding tissue; these granules appeared to be stained in the usual way. Other cells had a large ill-defined pale area in the centre and the granules were concentrated irregularly at one side of the cell. The smaller mast cells close to the blood vessels also had ruptured cell membranes. The total number of cells in the tissue had obviously diminished. The connective tissue cells showed increased basophilia and the connective tissue strands were visible as a basophilic network.

Ten days after irradiation mast cells could be seen as very pale ghost cells or had entirely disappeared in localized areas. Elsewhere the cell membranes had ruptured and the granules were scattered through the surrounding tissue (Pl. 1, fig. 1*b*). A few small deeply stained cells were visible along the blood vessels. These cells represented the commencement of recovery of the tissues from the effects of irradiation by the development of new cells from the adventitia of the blood cells (Riley & West, 1955*a*). The connective tissue network was still deeply stained with the basic dye. On the sixteenth day (Pl.1, fig. 1*c*) the mast cells consisted of a mixture of deeply stained small cells along the vessels and larger deeply stained cells in the tissue remote from the vessels.

A few cells with ruptured membranes and granules lying round them in the tissues, were still visible.

The mast cells of the stomach and jejunum. The mast cells in the submucosa of the stomach showed a series of changes similar to those in the subcutaneous tissue and occurring at similar times. Twenty-four hours after irradiation most of the cells were still normal. In some there was central pallor with concentration of the granules at one side, but none of the cell membranes were ruptured and there was no decrease in the number of cells (Pl. 1, fig. 2*a*). On the third post-irradiation day there were a few ruptured cell membranes and a few granules had leaked into the surrounding tissue. On the fifth post-irradiation day there was general rupturing of the cell membranes and granules were scattered through the surrounding tissue. Many of the cells showed vacuolation. On the tenth day (Pl. 1, fig. 2*b*), after irradiation the appearance of the cells was similar although a few deeply stained cells could now be seen beside the blood vessels. Very few mast cells could be seen in the wall of the jejunum of the normal rat. The cells became more difficult to find after irradiation, but in those which were seen it appeared that a sequence of changes occurred in them similar to that seen in the gastric wall.

The blood coagulation time. No gross increase in the coagulation time occurred during the 15 days following irradiation and the day-to-day variation which did occur was not significant (Text-fig. 5). This agrees with the observations of Cohn (1952) who found no change in the coagulation time in female rats following irradiation with 400 r.

Comparison of the different effects produced by irradiation. A comparison of the various effects produced by body irradiation with 1025 r is given in Table 1, which also includes results obtained by Cohn (1952) on the heparin coagulation time after body irradiation with 400 r. The effects on the histamine value of the skin, on the mast cells, and on the heparin coagulation time appeared at the same time after irradiation and reached their maximum during the following days. The effects on the histamine content of the alimentary canal, and on the epithelium lining the canal, appeared during the first twenty-four hours, but the latter were maximal several days before the histamine fell to its minimum value.

The effect of local irradiation of abdominal skin

In order to investigate the effect of doses of 1000 r on the abdominal skin histamine, the rats were partially protected by lead shields which diminished the mortality produced by the higher doses to the abdominal skin. Several days after irradiation of the abdominal skin its histamine content had fallen from the control value of 30·5 to 12·6 μg/g and then fell slightly further on the tenth post-irradiation day to 12·0 μg/g. The percentage effects of doses of 605 and 1000 r are shown in Text-fig. 6, in which it can be seen that the larger dose produced a greater fall in the skin histamine value, although the effect pro-

duced by each dose occurred after the same latent period. A dose of 2000 r from a circular applicator produced no changes in the histamine content of the abdominal skin 20 hr after local irradiation in comparison with adjacent unirradiated skin. Seven days after irradiation with 2000 r, the skin histamine values in the irradiated areas in two rats were 7·8 and 13·7 μg/g, whereas the histamine contents of adjacent unirradiated areas were 25·1 and 30·1 μg/g respectively.

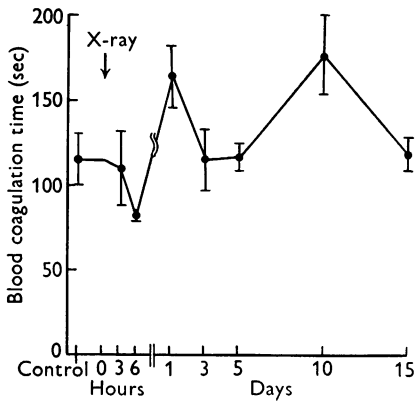


Fig. 5

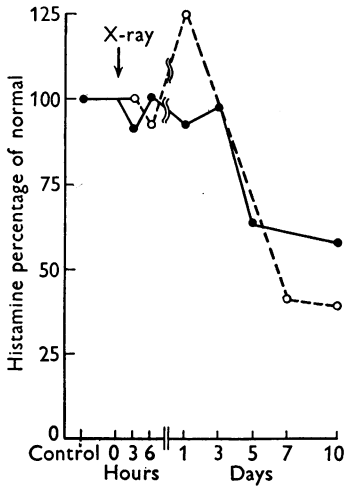


Fig. 6

Text-fig. 5. Effect of 1025 r total body X-irradiation applied to the skin of the back on the blood coagulation time. The standard error of the observations at each time interval is shown.

Text-fig. 6. The changes in the histamine content of the abdominal skin, following different doses of X-irradiation applied to the skin, expressed as percentages of the normal values. ●, 605 r; ○, 1000 r.

TABLE 1. Effect of total body irradiation on the histamine content and histological appearance of the tissues, and on the blood coagulation time

Tissue examined	Time of effect following irradiation: Onset	Time of effect following irradiation: Maximum	Maximum intensity
Skin, histamine	5 days	10 days	58% normal
Stomach, histamine	? 3 hr	5 days	58% normal
Jejunum, histamine	3 hr	5 days	8% normal
Skin, mast cells	5 days	10 days	Rupture of cell membranes, degranulation, decrease in numbers
Stomach, mast cells	5 days	10 days	
Stomach, mucous membrane	?	24 hr	Degeneration of all types of cells
Jejunum, mucous membrane	?	24 hr	
Blood, coagulation time	No effect	No effect	No effect
Blood, heparin coagulation time (Cohn, 1952)	6 days	16 days	25 × control values

Local reactions of skin vessels

Intradermal injections of either histamine or compound 48/80, at all concentrations tested, elicited about the same response in the normal skin and in the skin which had been irradiated locally 20 hr earlier with 2000 r. Thus at the time the response of the skin vessels to histamine and compound 48/80 was examined the histamine content in the skin had not yet decreased. Pontamine sky blue appeared in the normal and irradiated skin over approximately equal areas and with the same intensity (Pl. 1, fig. 3). In many rats the colour seemed to develop earlier at the irradiated site, but this difference soon vanished and was not noticeable by the tenth minute. Intradermal injections of saline produced only traces of local blueing.

DISCUSSION

It has been shown by Riley & West (1953) that a large portion of the histamine contained in the skin is present in the mast cells. In the present experiments degeneration of the mast cells in the subcutaneous tissue spreads appeared after a latent period of several days, at the time when the skin histamine value began to fall, and was most extensive 10 days after irradiation when the skin histamine was at its minimum value. As the cells in the subcutaneous tissue began to resume their normal appearance, so the skin histamine value began to rise again. These results resemble those described by Riley & West (1955*a*) following the injection of 'subacute doses' of compound 48/80 and suggest that the fall in the skin histamine is associated with the destruction of the mast cells in the skin. Similar degenerative changes have been reported after irradiation in the subcutaneous mast cells of the rat (Sylven, 1940) and in the cheek pouch of the hamster (Smith & Lewis, 1953). Increasing the dose of X-irradiation in the rat, which has been shown by Bloom (1948) to cause increased destruction of mast cells, produced a greater fall in the skin histamine.

Feldberg & Talesnik (1953) have shown that the alimentary canal is resistant to the action of the histamine liberators. That histamine can be held in the tissues independently of the mast cells is suggested by the fact that large doses of compound 48/80 and other liberators (Riley & West, 1955*b*; Brocklehurst, Humphrey & Perry, 1955) sufficient to cause complete destruction of the mast cells do not deplete the skin completely of histamine. In the jejunum and stomach the histamine values decreased several days before the gross changes in the mast cells occurred which Riley & West (1955*a*) report are correlated with changes in histamine content. It therefore appears that irradiation can deplete the alimentary canal of histamine independently of its action on mast cells, and thus it is suggested that the histamine in the upper part of the

alimentary canal may be held in the tissues of the wall independently of the mast cells.

It is claimed that one of the physiological functions of histamine in the gastric wall is the maintenance of the acid secretion by the parietal cells of the gastric mucosa (Code, 1956). A decrease in this acid secretion in human beings has been described by Palmer & Templeton (1939) following irradiation of the stomach. The mechanism by which irradiation causes this effect is unknown, but if irradiation causes a depletion of gastric histamine in man as it does in the rat, it is possible that the effect of irradiation on gastric secretion may be explicable in terms of its action on the histamine content of the stomach wall.

No characteristic signs of histamine release were observed at any time following irradiation. The signs of histamine release appear after the injection of a large dose of compound 48/80 which produces its effects on the mast cells within 3 hr (Riley & West, 1955*a*). The gradual decrease in tissue histamine which was observed in the present experiments may account for the absence of clinical signs of acute histamine release.

No significant change in the blood clotting time was observed at any stage. The finding by Cohn (1952), of a change in the heparin clotting time at the same time as a decrease in the skin histamine and degeneration of the mast cells following irradiation was observed in the present experiments, makes it probable that the mast cell degeneration determines both the changes in heparin clotting time and in the skin histamine.

Normal skin, and recently irradiated skin which still contained its normal histamine content, reacted to intradermal injection of histamine in the same way. This indicates that capillary permeability in these pieces of skin was similar (Miles & Miles, 1952). Compound 48/80 also caused intense local staining of the skin tissue by the dye to an equal degree in normal and irradiated skin. This effect is due to the local liberation of histamine, which is responsible for an increase in capillary permeability and consequent staining of the skin tissue (Feldberg & Miles, 1953). Therefore compound 48/80 was able to liberate sufficient histamine in both sites to cause equal changes in the capillaries, and the capillaries in both sites were still capable of reacting to the histamine to an equal degree.

Ungar & Damgaard (1954) have investigated the local skin reactions in rats to intradermal injections of histamine and compound 48/80 following β-radiation of the abdominal skin. They found that from 6 hr to 9 days after this treatment the normal skin response to compound 48/80 was abolished, although that to histamine remained the same as in normal skin. They investigated the local reactions by the observation of the wheals in the shaved abdominal skin, without the aid of a dye. It is unlikely that the different nature of the ionizing radiation is the cause of the discrepancy between the present results and those of Ungar & Damgaard (Warren, 1943; Prosser, 1947). In view of the fact that

compound 48/80 produces the skin reaction almost wholly by the local liberation of histamine, it appears unlikely that, provided there is sufficient histamine in the tissue, its actions should be different from those of histamine.

It has been shown in the present experiments that X-irradiation damaged the cells lining the stomach and jejunum. Cellular destruction of a similar nature has also been reported in these tissues by Pierce (1948) soon after exposure to radiation. These changes appeared at the same time as the tissue histamine began to decrease, but tissue repair was almost complete when the histamine content was still at its minimum.

The dry weights of the stomach and intestine decrease and the water content increases in rats between the first and ninth days following irradiation (Bowers & Scott, 1951*b*; Conard, 1952), but the percentage increase in water content is insufficient to account for the much larger percentage decrease in the histamine content of these tissues. A fall and then a rise takes place in the sodium and potassium contents of the stomach and intestine during the nine days following irradiation (Bowers & Scott, 1951*a*, *b*), but these changes are not correlated in any way with the steady decrease in histamine content. The protein content of the alimentary canal also diminishes following irradiation with neutrons at the same time and over the same period as the intestinal histamine decreased in our experiments (Ross & Ely, 1949). The changes in the histamine content following irradiation therefore appear to be related more closely to changes in the weight and protein content of the alimentary canal than to changes in its water or electrolyte content.

It has been shown that the administration of chloramphenicol to the rat can reduce the histamine content of the wall of the small intestine to about 35% of normal through its action on the histamine-forming bacteria of the intestine (Wilson, 1954). Irradiation causes changes in the bacterial flora of the caecum in the rat (Bell, Coniglio & Hudson, 1955), and it is thus possible that the changes in the present experiments might be attributable to the effect of irradiation on the bacterial flora of the intestine. However, this could not account for the changes observed in the gastric histamine content, and it is unlikely that it would affect any histamine-producing bacteria at the proximal end of the jejunum so grossly as to decrease the histamine in the wall of the jejunum by 90%. Exposure to X-irradiation is known to reduce the activity of cholinesterase in the rat intestine (Conard, 1952), and it is possible that it may influence the activity of the intestinal histaminase or histidine decarboxylase. In this way, by a primary effect on the enzymes responsible for the metabolism of histamine in the intestine, it may cause the observed changes in the intestinal histamine content.

Weber & Steggerda (1949) describe an increase in the plasma histamine concentration in rats during the first 2 hr and on the fifth day following irradiation, and Leitch & Haley (1955) state that a twofold increase in the

urinary excretion of histamine occurs in rats during the post-irradiation period. Venters & Painter (1950) have shown that in the rabbit and dog there is increased sensitivity to the infusion of histamine following irradiation, and have suggested that the radiation syndrome might partly be produced by the liberation of a toxic product from the mucous membrane of the small intestine. The sequence of the decreases in the tissue histamine which was observed in the present experiments, first in the alimentary canal and then in the skin, corresponds to the fluctuations in plasma histamine reported by Weber & Steggerda (1949) and provides an explanation for the source of the increased urinary excretion of histamine described by Leitch & Haley (1955).

SUMMARY

1. An examination was made in rats, during a period of 15 days following total body X-irradiation, of the histamine and mast cell content of abdominal skin, stomach and jejunum, of the blood coagulation time and of the histological changes in the epithelium lining the acid-secreting part of the stomach and in the jejunum.

2. Between the third and the fifth post-radiation days the skin histamine fell to 65% of normal and it remained at a low level during the remainder of the experiment. The subcutaneous mast cells did not show significant damage until the fifth day and the damage was more severe on the tenth day.

3. The histamine in the walls of the stomach and jejunum began to decrease during the first 24 hr and fell to minimum values of 58 and 8% of normal, respectively, after 5 days. These values were maintained throughout the rest of the experiment. The mucous membranes showed maximum damage after 24 hr and thereafter rapid repair occurred. The mast cells did not show degeneration until the fifth post-irradiation day.

4. No change occurred in the blood coagulation time throughout the experiment.

5. The appearance of pontamine sky blue in irradiated skin in response to intradermal injection of histamine or compound 48/80 20 hr after irradiation did not differ from that in normal skin, indicating that the capillaries react in the same way at both sites.

6. It is suggested that the fall in skin histamine is attributable to the concurrent degeneration of subcutaneous mast cells, but that the depletion of histamine in the alimentary canal is largely independent of changes in the mast cells.

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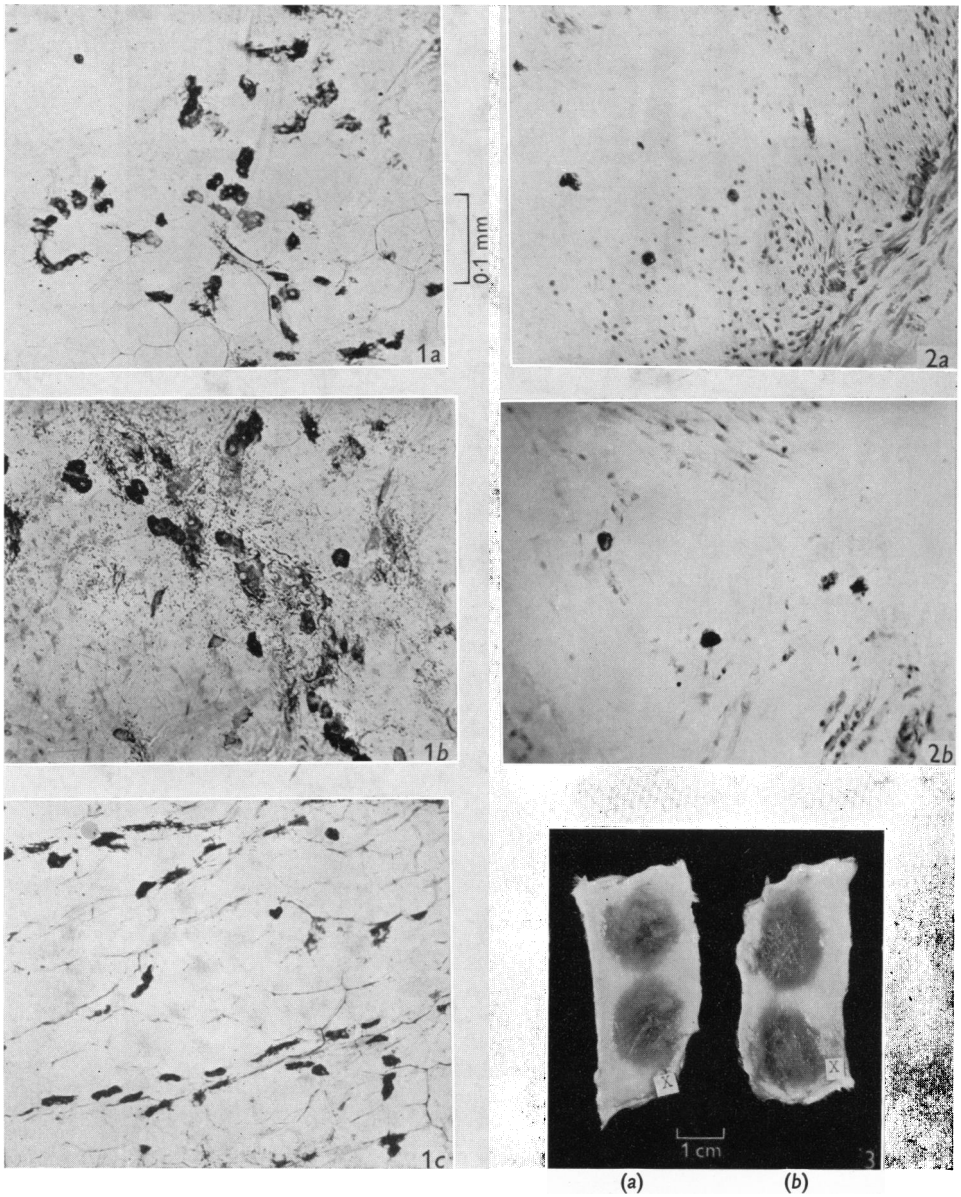

(*To face p.* 519)

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EXPLANATION OF PLATE

Fig. 1. Subcutaneous tissue spreads following 605 r X-irradiation to abdominal skin. Toluidine blue. (*a*) Fifth day following irradiation. Swelling of the mast cells and rupture of the cell membranes with dispersion of granules in the adjacent tissue. (*b*) Tenth day following irradiation. Swelling and extensive disintegration of the mast cells with wide dispersion of granules. (*c*) Fifteenth day following irradiation. Small new cell formation along the capillary at the bottom of the photograph. Disintegration of cells and dispersion of granules still visible at the top of the photograph.

Fig. 2. Section of the submucosa of the stomach following 1025 r total-body X-irradiation. Toluidine blue. (*a*) Twenty-four hours after irradiation. No significant changes in the histological appearance of the mast cells. (*b*) Tenth day after irradiation. Disruption of the cell membranes with dispersion of the granules in two cells, and vacuolation in another mast cell.

Fig. 3. Local skin reactions in X-irradiated and adjacent normal abdominal skin. X: irradiated areas. Blueing in response to intradermal injections of (*a*) Compound 48/80, 1/30,000; (*b*) histamine, 1/20,000.