HUMORAL EFFECTS OF METASTASISING CARCINOID TUMOURS

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Biörck, Axén, and Thorson (1952) and Thorson et al. (1954) drew attention to a curious syndrome found in some patients with malignant carcinoid tumour. The most constant feature is a peculiar type of flushing, but diarrhœa and right-sided valvular lesions of the heart may also be present. It has been suggested (Thorson et al. 1954) that the cause of the symptoms may be secretion of 5-hydroxytryptamine (serotonin) by the tumour, but biochemical observations are scanty. In this paper we present the clinical, pharmacological, and biochemical findings in three cases observed at the Manchester Royal Infirmary.

Case-records

PATIENT 1

At the age of 61, 9 months before his death, this patient first noticed unpleasant flushing of his face a few minutes after a drink of beer. When he next took beer, 6 weeks later, a single mouthful had the same effect, and 10 weeks after the original episode he began to have spontaneous attacks of flushing about twice a day. 5 months before his death, he had a gradually deepening permanent flush which grew deeper during periodic attacks; and these were associated with an urgent desire to defæcate. Some 4 months before his death, watery diarrhœa had become troublesome and his weight had fallen by one stone.

Investigations

When he was admitted, the skin of his face and neck was deep red and coarse, and his lips were cyanosed. The heart appeared normal. The liver was hard, irregular, and enlarged to the umbilicus. Chest radiography, barium meal, barium enema, full blood-count, electrocardiography, liver-function tests, and tests for occult blood in the stools showed nothing abnormal. A clinical diagnosis of malignant carcinoid was made, and this was supported by the finding of raised levels of 5-hydroxytryptamine (5-H.T.) in the blood, and 5-hydroxyindole-acetic-acid (5-H.I.A.A.) in the urine.

As little as 10 ml. of 10% alcohol, by mouth, or intravenously, caused an intense flush, which spread over the entire body within 5 min. An attack of flushing was accompanied by a rise in blood-pressure from 130/70 to 150/70 mm. Hg, tachycardia, and an uncomfortable sensation in the abdomen.

Progress of Illness

During the 5 months the patient was under observation the flushing rapidly became more intense until 1–2 months before his death. From that time, with increasing cachexia, the attacks faded; but the diarrhœa continued, and ankle œdema appeared and increased despite the use of mercurial diuretics. Finally the patient developed a respiratory infection and died. At no time were any cardiac murmurs heard. Consent for even a limited post-mortem examination was refused.

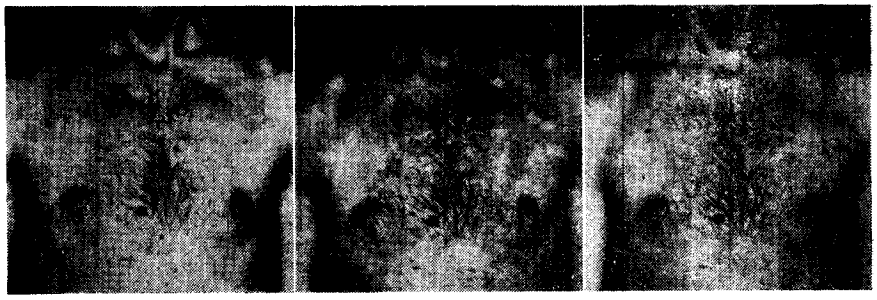
PATIENT 2

In a woman of 58, the illness began with watery diarrhœa, which continued until her death 2 years later. A month later, lethargy and weight loss were noticed, and after a further 9 months she developed progressive dyspnœa on exertion. 6 months before death she suddenly experienced a sensation of pressure in the epigastric region followed by unconsciousness lasting for 10 hours. This episode was repeated twelve times, nine of them occurring after admission 2 months before death. The attacks usually began with slight flushing of the face and hands; within a few minutes the patient became too weak to move. She complained of nausea, a sense of suffocation, and a feeling of severe epigastric pressure. The face and extremities became intensely cyanosed, while the rest of the skin was pale, cold, and moist; no peripheral pulsation could be felt even in the carotid arteries, but the heart sounds were normal, the rate being 100–120 per min. Despite this circulatory collapse consciousness was retained, except in the first attack. The retinal vessels showed no abnormality apart from cyanosis. The respiratory rate was only slightly increased; examination of the lungs revealed no cause for the feeling of suffocation. After several hours the patient gradually recovered, but she felt weak for a day or two. Occasionally the attacks were followed by œdema of the lips and hands. During one attack, in which an indwelling catheter was inserted, no urine was secreted for four hours.

From about the time of the first of these episodes, the patient also began to experience slight flushing of the face and hands up to three or four times a day. This flushing was usually but not always related to meals, and the only distressing feature was a forcible heart action. Often there was no change in her appearance, apart from the flushing, but transient blotches of cyanosis and pallor were observed once. Between attacks of flushing the patient felt moderately well, except for her persistent diarrhœa.

Investigations

The liver was firm and enlarged a hand-breadth below the costal margin. The blood-pressure was 130/90 mm. Hg, and a soft systolic murmur was audible at the base of the heart. Electrocardiograms (taken during and between episodes of collapse), chest radiography, barium meal, barium enema, a full blood-count, and liver-function tests showed no abnormality; a benzidine-test for occult-blood was positive. The taking of alcohol did not reproduce the symptoms. A clinical diagnosis of malignant carcinoid was made and was subsequently confirmed at necropsy.



A B C

Fig. 1—Flushing after alcohol (patient 3).

A, Before alcohol. B, Height of flush. C, Flush fading.

Operation

In view of the unimpressive results of medical treatment, and the improvement observed by Thorson et al. (1954) after the removal of a large mass of extrahepatic tumour tissue, laparotomy was undertaken (Mr. H. T. Simmons). The liver was found to be full of secondary deposits, some solid and others cystic, but no primary tumour could be found. With the induction of anæsthesia, the patient became flushed and respiration, which was being artificially assisted, offered considerably increased resistance, suggesting the presence of bronchospasm, although there was no evidence of this on auscultation. The blood-pressure rapidly became unrecordable and the extremities cyanosed. When the abdomen was opened the aorta was found to be pulsating poorly and could be easily compressed. The abdominal contents were not cyanosed. The patient regained consciousness after operation but the blood-pressure remained low, and after a few hours she lapsed into a deep coma, and became pulseless, cyanosed, and apnœic. Artificial respiration was carried out for several hours but she died 24 hours after operation.

Post-mortem Findings

At necropsy (Prof. A. C. P. Campbell), a smooth, grey primary tumour 1 cm. in diameter was found in a Meckel's diverticulum. The liver was enlarged (4 kg.) and full of secondary deposits, some pale grey and nodular, others cystic and containing bloodstained fluid. The only other secondary deposits were in two lymph-nodes in the mesentery and lesser omentum, and two small emboli of tumour cells in pulmonary arterioles.

Histologically all tumour-tissue was of a typical carcinoid structure. No mitoses were seen. The Masson-Fontana silver method showed brown argentaffin granules in the cytoplasm, especially in the cells at the peripheries of the masses. These granules also gave a positive reaction with the alkaline-diazo method (Pearse 1953). These reactions in the tumour cells were identical with those given by argentaffin cells in control sections of normal duodenum and appendix, thus confirming the tumour as an argentaffinoma.

No pulmonary stenosis was present, but the posterior cusp of the pulmonary-valve was slightly puckered by irregular fibrous thickening, and there was a smooth plaque of thickened intima in the pulmonary artery just above the anterior commissure.

PATIENT 3

A man aged 43 had had attacks of flushing for 20 years. These tended to be brought on by heavy meals, or even occasionally by a drink of water, and by emotion. They were always brought on by taking alcohol in any form. Neither the character nor the intensity of the flushing had changed over 20 years. 2 years before admission he began to suffer from episodes of nausea and vomiting, lasting 2 or 3 weeks, and occurring about every 3 months. Examination a year before admission had revealed a hard nodular liver extending a hand-breadth below the costal margin, suggesting the presence of secondary deposits. A barium meal showed no abnormality, and at no time had there been any diarrhœa.

Investigations

On admission the patient looked well. Weight loss over the preceding 3 years was only 10 lb. Apart from the enlarged liver, which had not changed since the year before, there was no physical abnormality. All investigations, including full blood-count, liver-function tests, cholecystography, and barium meal, were normal. It was found that flushing regularly began 4 minutes after taking 10 ml. of 10% alcohol, and lasted 15–20 min. The flush was accompanied by a rise in pulse-rate from 64 to 96 per min. and a rise in blood-pressure from 120/70 to 130/70 mm. Hg. There was no subjective sensation. A slightly more intense flush developed after the same dose of alcohol given intravenously. During an attack the conjunctivæ became suffused, and patches of erythema appeared over the chest, arms, and abdomen; patches of cyanosis appeared with the erythematous areas, and areas which had previously been pale became flushed as the attack subsided (fig. 1).

A clinical diagnosis of malignant carcinoid was made, and was supported by the finding of increased excretion of 5-H.I.A.A. in the urine.

Operation

Mr. Simmons performed a laparotomy in the hope of being able to resect the bulk of tumour tissue. The liver contained numerous large, white secondary deposits. A mass of glands was present along the lesser-curve of the stomach, and a small, white, submucous primary tumour 1·5 cm. in diameter was found on the anterior wall of the stomach. Since the glands accounted for only a small proportion of the tumour mass, it was not thought worth while attempting to resect them, but the primary tumour, one gland, and a secondary deposit in the liver were removed for histological examination. An incidental finding, which was thought to explain the attacks of nausea and vomiting, was the presence of an active duodenal ulcer.

The patient made an uneventful recovery and was sent home on a dietary régime. The flushing was at first unaffected by the operation, but when the patient was reviewed 4 months later, he stated that his flushings were less frequent and less severe, and two observers noted that the liver was softer and only extended two finger-breadths below the costal margin.

Histological Findings

Examination of the tumour and secondary deposits (Professor Campbell) showed solid alveolar masses of small epithelial cells with small round nuclei and pale eosinophilic cytoplasm deeply invading the muscle-coat of the stomach. The appearance was that of a carcinoid tumour, but there was more variation in the nuclear size and more tubular formation than is usual. No granules were demonstrated in the cells by using either the Masson-Fontana silver, or the alkaline-diazo methods.

Pharmacological and Biochemical Investigations

In 1953 Lembeck found a high content of 5-H.T. in carcinoid tumour tissue, and Thorson et al. (1954) suggested that this substance might be responsible for the syndrome. Up to the present, however, the only observations on this point have been limited to isolated serum 5-H.T. estimations on two patients exhibiting this syndrome (Pernow and Waldenstrom 1954), and estimates of 5-H.I.A.A. (a urinary excretion product of 5-H.T.) in three patients with malignant carcinoid, only one of whom was known to flush (Page et al. 1955). The evidence that 5-H.T. is responsible for the symptoms is therefore based on meagre evidence.

5-Hydroxytryptamine

In health virtually all circulating 5-hydroxytryptamine is contained in the platelets but less than half is found in

the serum after the blood has clotted (Hardisty and Stacey 1955a). When 5-H.T. is present in abnormal amounts this proportion may be different and serum estimates are not, therefore, a satisfactory index of whole-blood 5-H.T. values. More reliable information can be obtained by estimates on platelet-rich and platelet-poor plasma; this will also show whether or not the 5-H.T. free in the plasma is at its normal low value.

Acetone extracts of tissue, plasma, serum, urine, and cerebrospinal fluid were assayed on the atropinised rat uterus against a standard solution of 5-hydroxytryptamine creatinine-sulphate by the method described by Hardisty and Stacey (1955b), and the 5-H.T. concentration in whole blood calculated as described by them. This method of extraction and assay prevents interference by certain other smooth-muscle-contracting substances. Further evidence as to the nature of the active substances present was obtained by the use of lysergic acid diethylamide (L.S.D.) which in concentrations of 50 μg. per ml. is a specific antagonist on the rat uterus to tryptamine

TABLE I—5-HYDROXYTRYPTAMINE

(Results expressed as 5-H.T. base)

—	Case 1	Case 2	Case 3	Normal
Serum (μg. per ml.)	0·4	0·8–1·65	0·18–0·22	0·07 ±0·02 (25 subjects)
Blood (μg. per ml.)	0·5	1·15	0·45	0·16 ±0·06 (35 subjects)
Platelet-poor plasma (μg. per ml.)	0·05	0·3	0·16	Less than 0·02 (35 subjects)
Platelets (μg. per 10^9)	0·22	0·34	0·08	0·06 ±0·02 (35 subjects)
Urine (μg. per ml.)	0·65	0·4–20	11–12	Trace only
C.S.F. (μg. per ml.)	..	0·5	..	Less than 0·006 (5 subjects)
Tumour (μg. per g.) primary	..	..	0·32	..
Secondary ..	..	580 200 (necropsy)	0·08	..

and its derivatives (Gaddum 1953). This method would not differentiate between 5-H.T. and its *N*-methyl derivatives found in normal human urine by Bumpus and Page (1955). 5-H.I.A.A., excreted in large amounts by our patients, does not cause contraction of the rat uterus and 5-hydroxytryptophane, probably the immediate precursor of 5-H.T., has only about 10^{-5} the activity of 5-H.T. Errors due to the presence of these substances are, therefore, ruled out.

As table I shows, the active substance in all these extracts was completely antagonised by L.S.D., with the exception of that from the plasma and tumour tissue of patient 3.

Serum, blood, plasma, and platelets.—Only one estimation of the 5-H.T. in blood, plasma, and platelets was done in each case, those in patient 1 being obtained when he was moribund. One estimation on the serum was done on patient 1, three on patient 2, and four on patient 3. In all three patients, all fractions of blood 5-H.T. were raised, except the platelets in patient 3. The changes were greatest in patient 2, in whom the symptoms were most severe, and least in patient 3, in whom they were trivial. In patient 2 the 5-H.T. concentration in the serum in a random specimen was 0·8 μg. per ml., but towards the end of an episode of collapse it had risen to 1·65 μg. per ml. In patient 3, however, no significant change occurred in the serum 5-H.T. when a flush was provoked by alcohol (table III). Although these levels are considerably raised, they are much lower than the 12–15 μg. per ml. (5·2–6·5 μg. per ml. expressed as base) of 5-H.T. in serum reported by Pernow and Waldenstrom (1954).

Urine.—All three patients had considerable amounts of 5-H.T. in the urine—especially patient 3 whose estimates before, and 2 months after, operation did not differ significantly. The substance estimated was completely antagonised by L.S.D., although that found in the plasma and tumour tissue of the same patient was not.

TABLE II—5-HYDROXYINDOLE-ACETIC-ACID

—	Case 1	Case 2	Case 3	Normal
No. of observations ..	7	23	10	40
5-H.I.A.A. μg. per ml. ..	73–394	95–2020	6·8–44	1·1 – 5·1 (2·7 ±0·8)
5-H.I.A.A. mg. per g. creatinine	97–900	148–1230	22–46	1·0 – 6·6 (3·6 ±1·3)

Tumour tissue.—A high concentration of 5-H.T. (about a quarter of Lembeck's figure) was found in the secondary deposits removed at biopsy from patient 2; the primary tumour was too small to permit assay as well as histological examination. The amount found in the primary growth in patient 3 was much smaller, and the secondary deposit contained even less. In both specimens from this case some of the activity was not due to 5-H.T.

Cerebrospinal fluid.—Only one estimate was made, and this was on fluid obtained from patient 2 at necropsy. The fluid was heavily infected when extracted 9 days after death, and it is likely that considerable loss had occurred. However, large amounts of 5-H.T. were found.

5-Hydroxyindole-acetic-acid

As an alternative to the technically difficult serotonin assay, it has been suggested that estimation of 5-H.I.A.A. (a known urinary excretion product of 5-H.T.) might be a more convenient means of detecting excessive 5-H.T. secretion (*Lancet* 1954, Page et al. 1955).

Urinary 5-H.I.A.A. was estimated according to the method of Udenfriend et al. (1954). The range of values obtained for 5-H.I.A.A. in the three cases, expressed both in μg. per ml. and mg. per g. of creatinine are shown in table II.

All three patients had abnormally high 5-H.I.A.A. excretion, the output corresponding with the clinical severity of the disease and the blood 5-H.T. levels. The 5-H.I.A.A. levels are similar to those found by Page et al. (1955). The concentration reached 2 g. per litre in patient 2 on one occasion. In patient 3, in whom there was little day to day change in symptoms, there was only slight variation in output of 5-H.I.A.A., whereas in patients 1 and 2, both of whom had periodic exacerbations, the excretion fluctuated widely. Variation in the output in patient 2 of 5-H.I.A.A. is shown in fig. 2; a severe attack was immediately followed by a considerable increase in 5-H.I.A.A. excretion. A small rise in 5-H.I.A.A. excretion could also be detected in patient 3 after an alcohol-induced flush (table III).

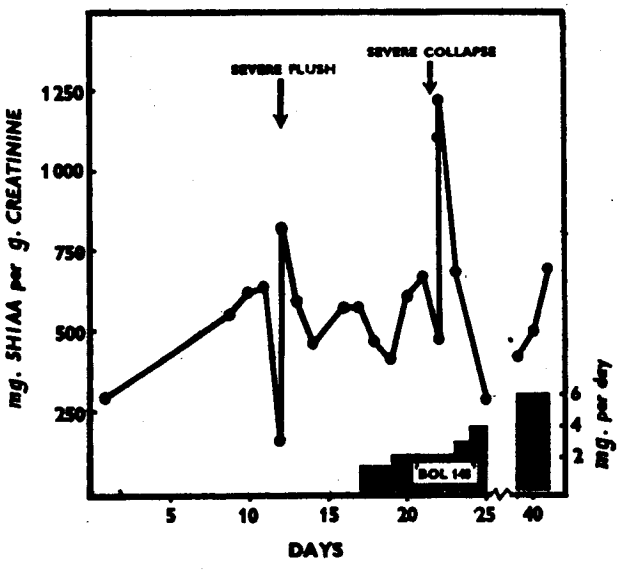


Fig. 2—Daily 5-H.I.A.A. excretion (patient 2).

Chromatography

Chromatography was used to confirm and extend the quantitative estimates of 5-H.T. and 5-H.I.A.A.

Ascending chromatograms were run for 12 hours at room-temperature. Two solvents were used: (1) *n*-butanol acetic-acid-water (4 : 1 : 5) for 5-H.I.A.A. and indole conjugates and (2) *n*-butanol-ethanol-2 N.ammonia (4 : 1 : 4) for 5-H.T. and methylated derivatives. After drying, the papers were sprayed with Ehrlich's solution (2% dimethyl-amino-benzaldehyde in 5% HCl), and heated to 50°C for 10–15 minutes to make them more sensitive than papers allowed to stand at room-temperature after spraying. Semi-quantitative estimations could be made by comparison of the intensity and area of spots with those caused by known amounts of the two substances (accuracy ± 30%).

Tumour.—Chromatograms in the second patient (fig. 4) indicated the presence of 0·4 mg. of 5-H.T. per g. of tumour tissue, which compares well with the pharmacological estimate of 0·58 mg. per g. No 5-H.I.A.A. could be found in the biopsy specimen, there was approximately 0·25 mg. per g. in the necropsy specimen, corresponding to a fall in 5-H.T. concentration from 0·58 mg. per g. before death to 0·20 mg. per g. after death (table I). No indole compounds could be demonstrated in concentrated tumour extracts from patient 3.

Urine.—5-H.T. was present in the urine of patients 1 and 3, the latter containing approximately 15 μg. per ml., which corresponds to the pharmacological estimate. 5-H.I.A.A. was present in the urine of all three patients (fig. 3) together with from 1 to 4 other indole spots, the most intense of which was identified as 5-H.I.A.A. sulphate ester.

TABLE III—INCREASED 5-H.I.A.A. EXCRETION AFTER FLUSHING (Patient 3)

—	Before alcohol		After alcohol (100 ml.)		
			30 min.	100 min.	180 min.
5-H.I.A.A. mg. per g. creatinine	24	27	40	30	30
Serum 5-H.T. μg. per ml. ..	..	0·21	0·22	0·18	..

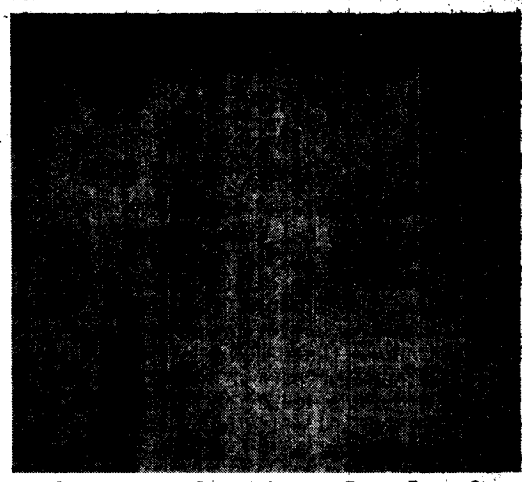
These spots had a low R_F in butanol acetic acid and did not give a strong colour reaction with diazotised sulphanilic acid, suggesting that they were 5-hydroxy indoles conjugated at the 5 position.

The major unknown indole, isolated by a method to be published elsewhere, had an R_F of 0·27 in butanol acetic acid and 0·20 in butanol-ethanol-ammonia. The ultraviolet absorption spectrum in N/5 NaOH was very similar to that of 5-H.I.A.A. On treatment with a limpet sulphatase preparation (Dodgson and Spencer 1953) it was broken down to 5-H.I.A.A., which was identified by chromatography in both the above solvents, and by colour reactions with Ehrlich and Pauli reagents. This indole thus appeared to be 5-H.I.A.A. sulphate ester. On paper electrophoresis (3hr., 200 V, 0·05 mA, pH 7·5, barbitone buffer) it moved towards the anode about twice as fast as 5-H.I.A.A. This is reasonable for the anion of a 5-H.I.A.A. derivative having two negative charges. Urines of rats given 10 mg. 5-H.T. intraperitoneally or 10 mg. 5-H.I.A.A. by mouth contained an indole with identical chromatographic and electrophoretic properties.

Hitherto, the only conjugated 5-hydroxy indole found to occur naturally is bufothionine (dehydro bufotenine sulphate ester) in the skin of some amphibia (Erspamer and Vialli 1952, Jensen 1935, Wieland and Vocke 1930).

Histamine

Pernow and Waldenstrom (1954) have reported "large amounts of histamine" in the urine of a patient with carcinomatosis from an unknown primary, who had attacks of flushing, and Feldberg and Smith (1953) have shown that 5-H.T. is a histamine liberator. We therefore studied the histamine content of blood and urine in our cases. Assays were kindly done for us by Mr. C. R. Beresford and Mr. P. Sheard of Bengers Ltd. Specimens of blood from all cases during flushing, and urine from patients 1 and 3 were taken by the method of Code (1937) and assayed on isolated atropinised guineapig ileum. No increase of histamine was found in any of the specimens.



A B C 5-H.I.A.A. D E F G

Fig. 3—Chromatograms of patients' urine compared with normal controls.

A, Patient 3. B, Patient 2 (after collapse). C, Patient 1. D–G, Normal controls. Marker 5-H.I.A.A. (5 μl. 0·01 M). (Solvent butanol acetic acid 40 μl. urine in each case.)

Effects of Treatment

In view of the possible rôle of 5-H.T. in the pathogenesis of the syndrome, the effects of various serotonin antagonists were tried.

Ergotamine tartrate.—Two doses of 0·25 and 0·5 mg. were given intravenously to the first patient, but without effect on the flush. The second patient received 0·25 mg. subcutaneously on two occasions before a meal, but the flush was not prevented.

Lysergic acid diethylamide.—This was given to the first patient on two successive days, in doses of 5 and 10 μg. This dosage caused an unpleasant throbbing noise in the head for several days, which precluded further increase in dosage; no effect was noted on the flushing or diarrhœa.

'*Bol 148*.'—This is a brominated derivative of L.S.D. (2 brom. *D*-lysergic acid diethylamide bitartrate), and is said by the makers to be about as potent as L.S.D. as an antagonist to 5-H.T., but without psychic side-effects. It was given to the second patient in a dose of 0·5 mg. daily working up to 7·5 mg. daily over a period of three weeks. There was no reduction in the number of flushes,

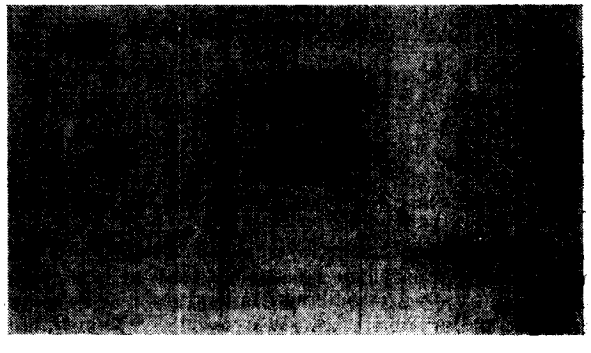


Fig. 4—Chromatogram of tumour extracts (patient 2) obtained post mortem, showing presence of 5-H.T. and 5-H.I.A.A. and absence of *n*-methyl 5-H.T. and bufotenine.

X. 100 μl. acid acetone extract of tumour (2 g. in 20 ml.)
X_1. Acid ether extract of X.
(Solvent butanol-ethanol-ammonia. Standard spots 5 μl. 0·005 M solution.)

but the patient thought that they were less severe. During treatment one severe attack occurred, which was indistinguishable from the previous ones. The daily excretion of 5-H.I.A.A. was unaltered (fig. 2). Patient 3 was given 0·5 mg. of Bol 148 every 2 hours for four doses, after which 10 ml. of 10% alcohol was given, causing a flush which was as intense and prolonged as previously.

Anti-histamines were tried empirically. The first patient was given an intravenous injection of 50 mg. of mepyramine, followed by 50 mg. intramuscularly 6-hourly for 6 days, without effect on the symptoms. The second patient was given 50 mg. of promethazine intramuscularly during each of the first three major attacks in hospital, but without improvement. 100 mg. of mepyramine orally three times daily for a week was also ineffective. The third patient was given 100 mg. of mepyramine by mouth hourly for 3 hours, followed by 10 ml. of 10% alcohol, which failed to cause the usual flush, but a further 30 ml. of 10% alcohol caused an intense flush, beginning after $5^1/_2$ minutes ($1^1/_2$ minutes later than usual). He also took three doses of 100 mg. of mepyramine during one day a week after his discharge from hospital, but he reported that this made no difference to his flushing.

We concluded that the 5-H.T. antagonists and antihistamines given in these doses were without benefit.

Surgery.—Thorson et al. (1954) were able to remove a large bulk of tumour tissue from one of their patients with subsequent relief of symptoms, presumably by removing a source of 5-H.T. In neither of our two patients who came to laparotomy was a large-scale resection possible. Resection of the primary tumour in patient 3 was not followed by any immediate change in the flushing, but the liver became smaller.

Discussion

Clinical Aspects

A notable feature of the cases collected by Thorson et al. (1954) was the variations in the duration of symptoms, ranging from a few months to 19 years, and Fraser (1955) has described a patient in whom symptoms began 36 years before death. A similar variation was present in our patients, one of whom died 9 months after the onset, and another is still alive despite having symptoms for 20 years. Some of these neoplasms are highly malignant and others are very slow growing, but in all cases of the syndrome so far described hepatic secondaries have been present. In our third patient, the small primary tumour contained very little active material, and the absence of any immediate change in the flushing after its removal suggests that the secondary deposits were mainly responsible for the symptoms. This poses the question whether or not metastases had been present for the whole 20 years during which flushing had occurred.

A second feature of our cases was the wide variation in the severity of the symptoms, ranging from simple flushing in patient 3 to complete collapse in patient 2. So far as we are aware, no such attacks as those occurring in the latter patient have previously been described in this syndrome, and the circulatory dynamics are not understood. Only patient 3 showed the geographical flushing described by Thorson and his colleagues, and even in this case patches of cyanosis as well as erythema were seldom seen. In the first patient, flushing was never patchy and apparently resembled that of the patient described by Branwood and Bain (1954). The patchy flushing, pallor, and cyanosis stressed by Thorson does not, therefore, appear to be constant.

Although the provocation of flushing by meals was mentioned by Thorson and his colleagues, extreme sensitivity to alcohol, as shown by patients 1 and 3, has not previously been described, and the mechanism is unknown. Acetic acid was without effect. Higher alcohols were not given.

The mental state of our patients was normal, despite the suggested rôle of 5-H.T. in mental processes (Woolley and Shaw 1954).

Pathogenesis of the Syndrome

There can be little doubt that some malignant carcinoids secrete 5-H.T., and it is reasonable to consider how far the symptoms found in our patients may be explained by the presence of large amounts of this substance in their blood. In comparing the symptoms with the reported pharmacological actions of 5-H.T., certain points should be borne in mind.

The 5-H.T. in the blood comprises two fractions; that bound in the platelets and that free in the plasma. The latter fraction is very small in health as platelets absorb 5-H.T. with avidity (Hardisty and Stacey 1955a) and free 5-H.T. is removed by the tissues (Erspamer 1954, Gaddum et al. 1953). The presence of large amounts free in the plasma as in patient 2 suggests that the eliminating mechanism was overloaded. It might be expected that free 5-H.T. would be more liable to cause symptoms than that bound in the platelets, and it would have been interesting to follow the changes in this fraction during a flush, had it been possible. Experience with isolated organs and with intravenous injections in animals has shown that tolerance is rapidly developed (Gaddum and Hameed 1954, Ginzel and Kottegoda 1954), and a patient who has had a high blood concentration of 5-H.T. for months or years may have very different signs and symptoms from a patient whose disease is of recent onset.

The effects of 5-H.T. in man have recently been studied (Spies and Stone 1952, Page and McCubbin 1953, Page et al. 1955). Instantaneous intravenous injections of 1·8 mg., or intravenous infusions of 5 mg. over 5 minutes, or 31 mg. over 60 minutes, caused unpleasant symptoms such as nausea, sensations of gasping for breath, tightness across the chest, desire to defæcate or micturate, and sometimes blushing and tingling. These symptoms mimic to a minor degree those experienced by our patient 2 during an episode of circulatory collapse. The characteristic flushing has not, however, been recorded after experimental injections.

Although 5-H.T. is a vasoconstrictor, its actions on the circulation are complex: in animal experiments hypertensive, hypotensive, or mixed responses are seen. Page et al. (1955) found only a small rise in mean arterial pressure (11–18 mm. Hg) in two normal human subjects in whom 5-H.T. was infused at a rate sufficient to cause unpleasant symptoms. The slight rise in blood-pressure during flushing in patients 1 and 3 and the episodes of circulatory collapse in patient 2 are not, therefore, inconsistent with the known actions of 5-H.T. The circulatory effects of 5-H.T. are not easily antagonised (Spies and Stone 1952, Woolley and Shaw 1955) and the failure of ergotamine tartrate, L.S.D., and Bol 148 is not surprising.

An unidentified active substance was found in the tumour tissue of patient 3, and, as there were differences between some clinical features of the syndrome and the effects of 5-H.T. given experimentally, it seemed possible that some substance other than 5-H.T. might be partly responsible for symptoms. It is known that 5-hydroxytryptophane is a precursor of 5-H.T. (Udenfriend et al. 1953, Clark et al. 1954) and that very small amounts of methylated serotonins are excreted in normal human urine (Bumpus and Page 1955). Chromatograms of tumour extracts and urine from our cases, however, did not reveal any of these substances.

In summary, large amounts of 5-H.T. were present in these three patients and there was a general relationship between the severity of the symptoms and the amount of 5-H.T. in the blood. We cannot at present exclude the presence of an active substance apart from 5-H.T. in these cases.

Diagnosis

The only constant clinical features of the syndrome which have so far been observed are flushing and hepatomegaly, and these findings alone may suggest the diagnosis. The addition of diarrhœa and valvular lesions of the right side of the heart confirm the diagnosis.

An increased excretion of 5-H.I.A.A. has been reported by Page et al. (1955) in a case of malignant carcinoid not showing these features, and by Clerc-Bory et al. (1954) in patients with other alimentary neoplasms. A small series of histologically or radiologically proved cases of carcinoma of the alimentary tract with hepatic secondaries studied by us showed a slightly raised 5-H.I.A.A. excretion ($5{\cdot}1 \pm 1{\cdot}9$ mg. per g. of creatinine) but these levels were much lower than those found in cases of malignant carcinoid with metastases. Blaschko and Hope (1955) have recently suggested that all urinary 5-H.I.A.A. may not be derived from 5-H.T., and that an increased excretion of 5-H.I.A.A. should be accepted as evidence of increased 5-H.T. metabolism with caution, Nevertheless, the daily output of 5-H.I.A.A. is probably at present the most valuable laboratory investigation in the diagnosis of malignant carcinoid.

It will not be possible to assess the value of 5-H.T. estimations in whole blood and blood fractions until we have more experience of the amount of free and platelet-bound 5-H.T. which may be found in these cases.

Summary

Three patients with flushing associated with abdominal carcinomatosis and liver metastases are described.

In the two cases from which material for histological examination was obtained, the tumours proved to be malignant carcinoids.

All three cases had abnormally high concentrations of 5-hydroxytryptamine in the blood, and 5-hydroxyindole-acetic-acid in the urine.

The clinical features of the syndrome and their pathogenesis are discussed.

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REFERENCES

Björck, G., Axén, O., Thorson, A. (1952) *Amer. Heart J.* **44**, 143.
Blaschko, H., Hope, D. B. (1955) *J. Physiol.* **129**, 11P.
Branwood, A. W., Bain, A. D. (1954) *Lancet*, ii, 1259.
Bumpus, F. M., Page, I. H. (1955) *J. biol. Chem.* **212**, 111.
Clark, C. T., Weissbach, H., Udenfriend, S. (1954) *Ibid*, **210**, 139.
Clerc-Bory, M., Pachéco, H., Mentzer, C. (1954) *C.R. Acad. Sci., Paris*, **238**, 525.
Code, C. F. (1937) *J. Physiol.* **89**, 257.
Dodgson, K. S., Spencer, B. (1953) *Biochem. J.* **55**, 315.
Erspamer, V. (1954) *Pharmacol. Rev.* **6**, 425.
— Vialli, M. (1952) *Ric. Sci.* **22**, 1420.
Feldberg, W., Smith, A. N. (1953) *Brit. J. Pharmacol.* **8**, 406.
Fraser, I. (1955) *Lancet*, ii, 174.
Gaddum, J. H. (1953) *J. Physiol.* **121**, 15P.
— Hameed, K. A. (1954) *Brit. J. Pharmacol.* **9**, 240.
— Hebb, C. O., Silver, A., Swan, A. A. B. (1953) *Quart. J. exp. Physiol.* **38**, 255.
Ginzel, K. H., Kottegoda, S. R. (1954) *J. Physiol.* **123**, 277.
Hardisty, R. M., Stacey, R. S. (1955a) *Ibid*, **129**, 24.
— — (1955b) *Ibid* (in the press).
Jensen, H. (1935) *J. Amer. chem. Soc.* **57**, 1765.
Lancet (1954) ii, 372.
Lembeck, F. (1953) *Nature, Lond.* **172**, 910.
Page, I. H., Corcoran, A. C., Udenfriend, S., Szoedsma, A., Weissbach, H. (1955) *Lancet*, i, 198.
— McCubbin, J. W. (1953) *Circulation Res.* **1**, 354.
Pearse, A. G. E. (1953) Histochemistry. London; p. 352.
Pernow, B., Waldenstrom, J. (1954) *Lancet*, ii, 951.
Spies, T. D., Stone, R. E. (1952) *J. Amer. med. Ass.* **150**, 1599.
Thorson, A., Björck, G., Björkman, G., Waldenstrom, J. (1954) *Amer. Heart J.* **47**, 795.
Udenfriend, S., Clark, C. T., Titus, E. O. (1953) *Fed. Proc.* **12**, 282.
— Titus, E. O., Weissbach, H. H. (1954) Personal communication.
Wieland, H., Vocke, F. (1930) *Liebigs Ann.* **481**, 215.
Woolley, D. W., Shaw, E. (1954) *Proc. nat. Acad. Sci., Wash.* **40**, 228.
— — (1955) *Fed. Proc.* **14**, 307.

CORRIGENDUM: ***Humoral Effects of Metastasising Carcinoid Tumours.***—In this article by Dr. P. J. D. Snow and his colleagues (Nov. 12, p. 1004) the third and fourth sentences of the second paragraph under "Chromatography" (p. 1007) should read:

"After drying, the papers were sprayed with Ehrlich's solution (2% dimethyl-amino-benzaldehyde in 5% HCl), and heated to 50°C for 10–15 minutes. The spots obtained in this way were usually more intense than those obtained when the paper was allowed to stand at room-temperature. Semi-quantitative estimations . . ."

Also under Chromatography, line 10 of the third paragraph of the section on urine should begin: "200 V, 0·05 M pH 7·5, barbitone buffer) . . ."