These doses of iron preparations are the *minimal* effective doses, and in resistant cases it may be necessary to double the dose here indicated. Colloidal and organic iron compounds are much less effective.

Vitamins

I usually augment this massive treatment with certain vitamin preparations such as bemax or marmite 1/2 ounce daily, or yeast compressed into 5-grain tablets. Some of these patients are almost certainly suffering from vitamin B deficiency, and I have had several cases which have improved upon these vitamin B containing preparations without any iron whatever. Infection, sepsis, pregnancy, and lactation all inhibit the response of these patients to iron, and in selected cases, where menorrhagia is present, artificial induction of the menopause may be necessary before improvement will take place.

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THE HAEMATOPOIETIC RESPONSE TO INTRAMUSCULAR INJECTIONS OF CONCENTRATED HUMAN GASTRIC JUICE

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Morris and his associates¹ recently reported that although small amounts of normal human gastric juice were ineffective when injected intramuscularly, large amounts of normal human gastric juice or hog gastric contents after concentration *in vacuo* produced maximal reticulocyte responses when injected into patients with pernicious anaemia. They described the substance in the concentrated gastric juice as being thermolabile, dialysable, and exhaustible, and therefore concluded that it was probably a hormone. Conner² reported similar results in one case of pernicious anaemia, following the injection of normal human gastric juice after concentration by vacuum distillation. Wilkinson³ was unable to confirm the findings of Morris and his associates when he injected human gastric juice that had been brought to pH 7.5 previous to its concentration *in vacuo*.

The observations here recorded were carried out in order to determine whether it was possible to separate the "active principle" in the gastric juice from the known enzymes (pepsin and rennin) by the process of ultrafiltration. This process not only offered a means of determining the relative size of the various constituents of the gastric juice, but also a possible method of concentrating the gastric juice at ice-box temperatures.

METHODS

The patients, clinically and haematologically typical of pernicious anaemia, received the concentrated gastric juice intramuscularly after preliminary control periods (average seven days). Red blood counts, haemoglobin determinations (Newcomer), and reticulocyte counts were made daily during the control and test periods. The human gastric juice used was obtained from young healthy adults following the injection of 0.5 mg. of histamine subcutaneously. It was filtered immediately after collection through a No. 1117½ Faltenfilter (Carl Schleicher and Schüll). The enzymatic content of the gastric juice to be concentrated was determined by methods detailed previously.⁴

The Bechold type of ultrafilters described by Bronfenbrenner⁵ was used in the experiments. The gastric juice (285 to 630 c.cm.) was concentrated by these ultrafilters, and twice diluted with distilled water or 0.3 per cent. hydrochloric acid, and reconcentrated before being brought to the final volume of 13 to 15 c.cm. This procedure was carried on in the ice-box. Since only negligible amounts of pepsin and rennin passed through these filters, each new filter could be tested for possible defects in the construction by determining the amounts of these known enzymes in the ultrafiltrate.

In a preliminary study it was found that the "active principle" in liver extract No. 343 passed through the ultrafilters. One patient (Case 9) had a reticulocyte peak of 25.6 per cent. (absolute reticulocyte count 620,000 per c.mm.) following the intravenous injection of the amount of liver extract derived from 100 grams of whole liver, after the extract had been passed through the ultrafilter.

In the experiments in which concentration by vacuum distillation was employed the procedure was as follows. The distilling flask (fitted with the usual capillary tube to prevent bumping) was immersed in a constant temperature bath maintained at 40°C. The vacuum was produced by a water pump. The gastric juice (500 to 600 c.cm.) was concentrated to 10 to 15 c.cm. in a period of three to five hours. The gastric juice concentrated by either or both of the above methods was sterilized by passing through an N Berkefeld filter, or by the addition of a few drops of tricresol, and was adjusted to a pH 7 (at ice-box temperature) by the addition of strong NaOH before being injected into the patients.

RESULTS

In the first group of observations (Group 1) the gastric juice used had been filtered through paper and then stored in the ice-box for approximately two months before being subjected to ultrafiltration. The effect of the two injections of the concentrated juice (that held back by the ultrafilters) is shown in Chart I, Case 1. The clinical response in the patient was only slight, and the increases in the red blood count were not sustained. A further rise in reticulocytes and a satisfactory rise in red blood cells followed the administration of extralin* by the mouth. The ultrafiltrate (the portion passing through the ultrafilters) of this juice was concentrated by vacuum distillation and injected into Case 2a. No response of the blood occurred during the following twelve days.

These reticulocyte responses were not maximal; therefore it was decided that fresh gastric juice should be used in the next group of tests (Group 2), as the storage in the ice-box might have destroyed some of the active material. However, when the fresh gastric juice was used, no reticulocyte responses followed the injection on three occasions of the portion of the fresh gastric juice held back by the ultrafilters, and on one occasion

* A potent preparation obtained by the interaction of liver or liver extract with hog gastric tissue.

the ultrafiltrate after it was concentrated by vacuum distillation. Chart II (Case 4) is an example of the results in this group.

Morris and his associates reported maximal responses of the reticulocytes following the injection of similar amounts of gastric juice after concentration by vacuum distillation, and yet we obtained only slight responses

CHART I.—The response of the blood of Case 1 to two intramuscular injections of human gastric juice that had been stored in the ice-box for two months before being concentrated by ultrafiltration, and to the oral administration of extralin capsules 4, three times a day.

when the gastric juice which had been stored in the ice-box for two months was concentrated by ultrafiltration and injected, and no responses when the fresh gastric juice was concentrated by ultrafiltration and injected. To test whether the concentration by vacuum distillation

had caused some change in the fresh gastric juice, it was decided to subject the latter to concentration by vacuum distillation before using the ultrafilters (Group 3). Case 6 was injected with gastric juice concentrated by vacuum distillation only. A maximum reticulocyte response (11.2 per cent. at a red blood cell level of 1.82 millions), and a satisfactory rise in red blood cells followed. No further reticulocyte rise followed extralin therapy. Four patients received injections of gastric juice first concentrated by vacuum distillation, then diluted with distilled water, and reconcentrated by ultrafiltration. Definite reticulocyte rises followed all of these injections. The reticulocyte rises were 9.8, 8.7, 8.2, and 2.8 per cent. when the red blood cell levels were 0.7, 3.25, 1.91, and 2.7 millions, respectively. The reticulocyte peak of 8.7 per cent. in Case 8 was apparently maximal, as no further rise in reticulocytes followed the injection of liver extract intramuscularly.

CHART II.—The response of the blood of Case 4 to the intramuscular injection of fresh human gastric juice concentrated by ultrafiltration only, and to the weekly intravenous injection of the amount of liver extract derived from 100 grams of whole liver.

Case 11 had previously responded to the concentrated ultrafiltrate, and the 2.8 per cent. response, followed by a rise in red blood cells, was considered quite satisfactory. The two other responses were not maximal, as subsequent treatment produced additional reticulocytoses.

Case 11 received intramuscularly the portion of the gastric juice concentrated by vacuum distillation that

CHART III.—This shows the response of the blood of Case 11 to the intramuscular injection of the portion of the fresh human gastric juice concentrated by vacuum distillation that passed through the ultrafilter (1), and to the portion of the fresh human gastric juice concentrated by vacuum distillation and then reconcentrated by ultrafiltration (2).

passed through the ultrafilters. This ultrafiltrate was concentrated by vacuum distillation before injection. Chart III shows the marked and prolonged reticulocytosis. Evidence of bone marrow stimulation (many nucleated

red blood cells, myelocytes, and nuclear particles) appeared at the onset of the greatest rise in reticulocytes. Case 7, in which there had been nine previous responses to liver extract, received a similar preparation. The patient was in cardiac failure when injected. By the seventh day the reticulocytes had started to increase, but the cardiac failure had not responded to medication, and the patient developed bronchopneumonia and died. After each injection in this group there was some type of reaction, the severest

following the injection (Case 11, first injection) which caused the greatest reticulocytosis. The clinical response of each patient was proportionate to the height of the reticulocyte responses.

The results in the three groups of observations indicated clearly that some change in the gastric juice had taken place during the process of vacuum distillation. However, in Group 2 the ultrafiltrate of fresh gastric juice had been

concentrated by vacuum distillation, and no haematopoietic activity had been demonstrated. It was therefore assumed that the material in the gastric juice that could be made active by vacuum distillation was held back by the ultrafilters. To test this, Case 12 was injected with fresh gastric juice first concentrated by ultrafiltration, then diluted with 0.3 per cent. hydrochloric acid, and re-concentrated by vacuum distillation. The patient had a severe reaction, a decrease in red blood cells, and an increase in icterus following the injection. Chart IV

CHART IV.—This shows the prolonged reticulocyte response of Case 12 following the intramuscular injection of normal human gastric juice first concentrated by ultrafiltration, then diluted and re-concentrated by vacuum distillation, and the response of the blood to three intramuscular injections of the amount of concentrated liver extract (3 c.cm.) derived from 100 grams of whole liver.

shows the prolonged reticulocytosis, the peak (345,000 per c.mm.) being reached on the thirty-fifth day after the injection. A definite clinical response and rise in red blood cells had occurred by then, although the rise in red blood cells was markedly delayed. There was a subsequent increase in reticulocytes following the intramuscular injection of liver extract.

This showed conclusively that the material in the gastric juice which could be made active was held back by the ultrafilters used. It also demonstrated that the gastric juice concentrated by ultrafiltration and re-concentrated by vacuum distillation was haematopoietically active, whereas on three occasions (Group 2) the gastric juice concentrated by ultrafiltration only had been inactive. The results obtained in Group 3 and in Case 12 indicated that after concentration by vacuum distillation a goodly percentage of the activity passed through the ultrafilters, although practically all of the material that could be released or made active by concentration by vacuum distillation was held back by the ultrafilters. It would seem, therefore, that during the process of vacuum distillation a haematopoietically active substance was obtained that was smaller in size than the material from which it was formed or released.

SUMMARY

The results of these studies indicate that some change in fresh human gastric juice must take place before a haematopoietically active material can be demonstrated by the intramuscular injection into patients with pernicious anaemia. At the present time the actual mechanism of this change is not known. Any of at least three explanations might account for this change. The first is that, during the process of vacuum distillation the intrinsic factor acts upon an extrinsic factor present

in the gastric juice in too small amounts to be active when fed by the mouth. The intrinsic factor is known to be present in fresh human gastric juice, and has been demonstrated by us in another group of experiments to be present at the onset of the vacuum distillation in all the preparations which were made active by this procedure, and in none of those that were not so made active. The second is that during the process of concentration by vacuum distillation, a material irritating or toxic to the haematopoietic system is produced. The prolonged and delayed reticulocyte response, marked bone marrow irritation, the delayed rise in red blood cells, and the fact that each patient who responded to the injection of the concentrated gastric juice had a more or less severe reaction, suggest this possibility. Case 12 is of especial interest in this connexion. After the injection the temperature rose to 106° F., the red blood cell count decreased 0.55 million, and there was an increase in icterus. There was no rise in red blood cells until after a prolonged reticulocytosis. The third possibility is that during the concentration by vacuum distillation a hormone is released or activated.

In our opinion the haematopoietically active substance must be formed by the action of the intrinsic factor on an extrinsic factor in the gastric juice, or by the production of a substance irritating or toxic to the haematopoietic system.

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ADDITIONAL SYMPTOMATOLOGY IN SIMPLE ACHLORHYDRIC ANAEMIA

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Although its recognition has been slow, its nomenclature varied, and its features diverse, unequal, and inconstant, a type of non-haemolytic primary anaemia, associated with a relative gastric achlorhydria, has of recent years been admitted a separate clinical entity since its distinctive pathology was made clear by Witts¹ in the Goulstonian Lectures of 1932. The object of this communication is to indicate two additional features of the symptomatology, believed not to have been hitherto recorded, which occurred in patients admitted to St. Thomas's Hospital Medical Unit during 1932. These are: (1) acne rosacea, and (2) thinning of the hair. It must be admitted in each case that a single example of such a symptom might appear at first sight to have occurred fortuitously, in spite of the diversity of symptoms—more especially abdominal—which are known to occur; but it is considered justifiable to record them, if only because they might be expected on pathological grounds.

ACNE ROSACEA

The skin condition was observed in a woman with most of the usual features of simple achlorhydric anaemia.

Mrs. G., aged 60, complained of palpitations and shortness of breath for six months, accompanied by pain in the epigastrium, chiefly post-prandial. She had long come to