CLINICAL PATHOLOGY IN GENERAL PRACTICE**LABORATORY INVESTIGATIONS IN THE DIAGNOSIS OF IRON-DEFICIENCY AND SIMPLE CHRONIC ANAEMIA**

BY

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Iron-deficiency anaemia occurs when the body gains iron more slowly than it should to provide for the needs of growth or to compensate for loss by haemorrhage. Only this factor of iron balance is of importance, so that nutrition enters into consideration only in so far as the food contains iron. Once iron deficiency has been demonstrated the laboratory tests to be used will be those which detect imperfect absorption of iron or loss of iron—that is, bleeding. It is easy to demonstrate iron deficiency, but not so easy to demonstrate its precise aetiology. The diagnosis of iron deficiency is complete only when the relative importance of growth, iron absorption, and blood loss have been assessed and when the site of blood loss is known. Every pathologist has performed necropsies on patients with carcinoma of the colon who had been treated for months for their iron-deficiency anaemia—treated without investigation until their resectable carcinoma had metastasized. It would be, of course, impossible to investigate exhaustively every patient, so the choice of investigation must depend on knowledge of the natural history of iron-deficiency anaemia.

Iron deficiency occurs during the first six months of life in premature infants, whose parasitism on their mother is cut short before they have obtained all the calcium and iron they need: the anaemia is more severe the more premature the birth. Childhood is usually free from iron deficiency, unless nose-bleeds are very severe, or bleeding occurs from peptic ulcer in a Meckel's diverticulum, or a peptic ulcer develops in a hiatus hernia. The menarche may lead after a few years to chlorosis in a young woman whose dietary fads are more extreme than usual; each pregnancy costs the mother at least 300 mg. of iron—a year's absorption from food (and bleeding from peptic ulcer in a hiatus hernia is commoner in pregnant women than in other subjects); and an abortion may cause even greater losses. In the fourth and fifth decades fibroids cause menorrhagia and absorption of iron is less because achlorhydria becomes common. Throughout adult life, in men as well as women, peptic ulceration and piles contribute to anaemia, and from the fifth decade malignant disease of the gastro-intestinal tract must always be considered. There are other causes of blood loss, such as ulcerative colitis, Crohn's regional ileitis, diverticulitis, and tuberculous ileitis, but in these anaemia is rarely the presenting symptom.

Iron deficiency is usually accompanied by some reduction in haemoglobin level, to 12.4 g. (80%) or below, but one occasionally sees anaemic polycythaemia with haemoglobin levels of 100% (14.8 g.) or more, though iron deficient.

Recognition of Iron Deficiency

Estimation of haemoglobin and of packed cell volume allows the calculation of the mean corpuscular haemoglobin concentration (see last week's article in this series, February 6, p. 326); if below 30% this indicates iron deficiency, between 30 and 32% iron deficiency may be present, but it is slight, while over 32% it is absent. The

stained film will show anisochromia, and if the anaemia is moderate or severe the cells will tend to be small, misshapen, and to vary excessively in size (microcytosis, poikilocytosis, anisocytosis) while they will contain less haemoglobin than normal (hypochromia). In severe anaemia target cells are often present, and they may lead to confusion with thalassaemia (Mediterranean anaemia or trait), found in Cypriots, Italians, and Greeks, or sickle-cell anaemia or trait, found in Africans and a few tribes of India or Persia, in neither of which congenital peculiarities does iron have any therapeutic value. If it is possible to estimate serum iron, it will be found to be lowered in iron deficiency and normal in the congenital anaemias.

Having established the presence of iron deficiency, one must consider aetiology; and if the history given by the patient clearly indicates a simple cause, such as menorrhagia or addiction to eccentric foods, treatment can be started without delay. If, however, there are no leading symptoms or signs one must first exclude bleeding from the gut.

Gregerson's benzidine slide test is here very useful. Powders of 25 mg. of benzidine and 200 mg. of barium peroxide are dissolved in 5 ml. of glacial acetic acid; a smear of faeces is made on a slide and two or three drops of the reagent added; a blue colour appearing within 30 seconds is presumptive evidence of blood, though this should be confirmed by the standard benzidine or orthotolidine tests after the patient has been on a meat-free diet for three to four days. A positive Gregerson test is not proof of bleeding, but it establishes a *prima facie* case for further investigation. One should, of course, exclude the presence of bleeding piles before using it, and this necessitates proctoscopy, while in older persons a digital examination may reveal a carcinoma.

Refractory Iron Deficiency

Once the diagnosis is made and the patient given iron and ammonium citrate mixture or ferrous sulphate compound tablets improvement should be rapid: the haemoglobin should rise by 4–20% weekly. If this rise fails to occur, it may be because the patient is not taking the iron or because it is not being absorbed.

Patients, especially women, often fail to take iron tablets because they claim that the tablets give them indigestion, diarrhoea, or constipation. This can be avoided by instructing them to take for one week one tablet in the middle of the main meal of the day; in the second week to add a second tablet in the middle of the next meal; and, thereafter, a third tablet in the middle of the third meal. Alternatively one of the preparations of ferrous gluconate may be prescribed, for there is no doubt that these cause less digestive disturbances than does ferrous sulphate. However, some patients take their tablets religiously without in any way affecting their anaemia, and this is an indication for the administration of iron by vein; it is also an indication for examination of the stools, for many patients who fail to respond to iron by mouth can be shown to have steatorrhoea. The classical method of Holt, Courtenay, and Fales for the estimation of faecal fat, which was performed on an isolated dried specimen, is now less used and is being replaced by methods in which an aliquot of the 24-hour stool is analysed without drying. This change in method reflects the interest in fat balance rather than fat excretion. To-day one expects over 90% of ingested fat to be absorbed and but 10% excreted; any greater excretion is considered evidence of steatorrhoea. If steatorrhoea is present, iron absorption virtually ceases, and steatorrhoea is the com-

monest cause of the finding that an iron-deficiency anaemia fails to respond to iron by mouth but responds rapidly to intravenous iron. Steatorrhoea causes iron deficiency far more often than it does megaloblastic anaemia.

Anaemias Secondary to Other Diseases

The production of red cells may be slowed or their destruction made more rapid by a number of diseases which have no direct association with the blood or bone marrow. Red-cell production is diminished by infection, by nitrogen retention (chronic uraemia) as in chronic nephritis, and by other disease states such as rheumatoid arthritis and myxoedema. That an anaemia may be due to one of these causes should be suspected when a steady or slowly progressing anaemia, usually of moderate severity (haemoglobin over 10 g.) is not due to iron deficiency (normal M.C.H.C.), and the cell volume is near or slightly below the lower limit of normal (M.C.V. 70–85 μ^3), though in myxoedema the cells may be slightly enlarged (macrocytosis, M.C.V. 95–115 μ^3).

In all such anaemias, if the blood count gives no clear indication for therapy a series of investigations may be initiated, each designed to exclude one or other cause. Adequate history-taking and physical examination should exclude the grosser forms of disease such as bronchiectasis, diverticulitis, osteomyelitis, or ulcerative colitis, but a differential leucocyte count showing neutrophilia and a raised erythrocyte sedimentation rate would suggest that a further and more intensive search for a site of infection might be desirable. Some infective processes show minimal changes in leucocyte counts, and of these tonsillitis and subacute bacterial endocarditis are very important; in these the count may be quite normal or there may be slight neutrophilia and lymphocytopenia, usually with some monocytosis, though such changes are inconstant. In typhoid fever (though not in the paratyphoids) and in brucellosis there are neutropenia and monocytosis. Brucellosis occasionally becomes very chronic, causing anaemia with only occasional bouts of fever; in such patients the monocytes may be mistaken for lymphocytes and a diagnosis of lymphatic leukaemia entertained. Fortunately, brucellosis is fairly uncommon in Britain, but agglutination reactions may be worth performing in a patient with an obscure anaemia and some fever. Pulmonary tuberculosis is a much more common cause of moderate anaemia, especially in young adults, and now that facilities for chest radiography are available almost everywhere it should be recognized easily.

An important cause of simple chronic anaemia is nitrogen retention, chronic uraemia. If a patient has a very slowly progressing renal lesion he may have no complaints until his blood urea exceeds 100 mg. per 100 ml., and his haemoglobin has sunk to, say, 60%. In such cases the haemoglobin level and blood urea are reciprocally related, so that if the blood urea can be lowered the haemoglobin level will rise. Such patients usually have an acidosis secondary to the renal lesion, and if this is present, as shown by a diminished alkali reserve (plasma bicarbonate), then administration of alkali (sodium bicarbonate) in quantities sufficient to raise the alkali reserve to, and to maintain it at, the middle of the normal range (68 vols. per 100 ml. or 30 mEq per litre) will result in a reduction in the blood urea and an increase in haemoglobin level. By this means a patient with nitrogen retention due to polycystic kidneys may be kept alive and useful for several years after he would have died from uraemia had he not been treated. This may be of great importance to the family of such a man, in whom death usually occurs during the fifth decade, when his children still need him; it is equally important for the child or young adult with renal rickets, whose bony lesion may be healed by precisely the same procedure. It is always desirable to estimate the blood urea in a patient with unexplained simple chronic anaemia, and, if it is raised, examination of the urine for cells and casts, careful palpation of the abdomen for polycystic kidneys, adjustment of the alkali reserve to the optimal figure, and perhaps an excretion pyelogram may be reasonable procedures.

There are other unrelated conditions which diminish red-cell production: some simulate an infective process—for example, rheumatoid arthritis; others appear to be neoplastic, such as Hodgkin's disease and other reticuloses, but in these the anaemia is rarely the presenting symptom, as it is often in uraemia and sometimes in infection, rather is it a symptomatic epiphenomenon. Occasionally one encounters a simple chronic anaemia in an obese woman, usually slow witted, who feels the cold very severely, who snores or who speaks slowly with a nasal intonation, and whose hair and eyebrows are greying and scanty. In such patients one can but suspect myxoedema, and a raised serum cholesterol would support that diagnosis and warrant further investigation, including a B.M.R., though photographs before and after a month's course of thyroid extract offer more convincing diagnostic evidence than does the B.M.R. One form of myxoedema can be caused by absorption of resorcinol from ointments or suppositories containing it, usually in middle-aged women with varicose ulcers.

Drug-induced chronic anaemia or chronic anaemia resulting from poisoning are both rare. Benzene inhaled from rubber solutions, dry-cleaning fluids, paints and varnishes, motor spirit, or from solvents may cause a hypoplastic anaemia, with leucopenia and thrombocytopenia developing during exposure or months or years thereafter, but this is rare in Great Britain. A similar anaemia may develop after the administration of chloramphenicol, though little is known of its frequency. Lead-poisoning is usually accompanied by a moderate anaemia with punctate basophilia, but should not be diagnosed merely because the blood cells show excessive punctate basophilia; one cannot say that a patient has been poisoned unless he develops symptoms or suffers organic damage. Not rarely a subject absorbs and excretes some lead without developing any symptoms, and if on the discovery of punctate basophilia he is said to have lead-poisoning he may well develop a compensation neurosis and manufacture symptoms. If lead absorption or even poisoning is suspected from the blood count, one should not suggest that the patient is "suffering from lead-poisoning" but should suggest that, "since he has been exposed to lead, it would be wise to discover how much he has absorbed" and arrange for an analysis of a 24-hour specimen of urine. If he has wrist-drop or abdominal colic and his lead excretion is excessive one has no alternative but to make the diagnosis of poisoning.

Unusual Blood States

There remain a few conditions which should be regarded as physiological aberrations, or necessary consequences of disease. Chronic retention of carbon dioxide is found in emphysema, and is accompanied by an increase in mean cell diameter up to 8.8 μ without increase in variability—a change also seen occasionally in congestive cardiac failure.

Unexplained macrocytosis without anisocytosis and without anaemia is rare and may be ignored if haemolytic anaemia and carcinomatous deposits in the bone marrow can be excluded. In obstructive jaundice the red cells become excessively thin and target cells are common, even though there is no iron deficiency; rarely, obstructive jaundice can thin the cells of acholuric jaundice (congenital spherocytosis) so much that fragility becomes normal. Occasionally, ovalocytosis, in which the cells are ellipsoidal disks, may be seen and must be differentiated from pernicious anaemia; in ovalocytosis anaemia is slight or absent and anisocytosis minimal. If necessary, a sternal puncture would show megaloblastic erythropoiesis in pernicious anaemia and normoblastic erythropoiesis in ovalocytosis.

It may be noted that no anaemia has been ascribed to vitamin deficiency. In Britain to-day such anaemias are so rare that no haematologist would consider such a diagnosis without a clear history of dietary deficiency and the symptoms of scurvy, beriberi, or pellagra. On the other hand, dysphagia with stomatitis, the so-called "Plummer-Vinson syndrome," may occur in elderly women who are not anaemic, and is then due to riboflavine deficiency.