

Juvenile Pernicious Anaemia

S. B. DIMSON

From the Children's Hospital, Sydenham, London, S.E.26

The extreme rarity of pernicious anaemia in childhood has been stressed by Lambert, Pranker, and Smellie (1961) who, by insisting on positive evidence of intrinsic factor deficiency for the diagnosis, could accept unequivocally only 7 cases in the literature but added 2 of their own, who were sibs. They may have overlooked the case described by Wilkins (1957) which was later confirmed by McIntyre, Hahn, Conley, and Glass (1959). Since then, Leikin (1960) and Waters and Murphy (1963) have each reported 2 cases, also in sibs, while Morse, Cochrane, and Landrigan (1961), Clement, Nichol, and Welch (1961), Hung, Migeon, and Parrott (1963), and Pearson, Vinson, and Smith (1964) have added one each making the total 18.

However, the cases of Reisner and Ellsworth (1955), Wilkins (1957), Morse *et al.* (1961), and Hung *et al.* (1963) were complicated by idiopathic hypoparathyroidism which usually preceded pernicious anaemia by 5 to 7 years. Since hypocalcaemia is thought to depress gastric function (Donegan and Spiro, 1960) and possibly also the secretion of intrinsic factor (Waters and Murphy, 1963), cases following endocrine disorders should be treated with reserve when conclusions are drawn regarding the nature of juvenile pernicious anaemia.

A condition which is very similar clinically and which must be distinguished from juvenile pernicious anaemia is a chronic relapsing megaloblastic anaemia in children whose intrinsic factor activity is normal and who show no evidence of malabsorption. Imerslund (1960) described 10 such cases in 6 families. All showed a low serum vitamin B12 level but had a normal vitamin B12-binding factor in their gastric juice. In addition, persistent proteinuria and urinary tract abnormalities were common. Imerslund concluded that the megaloblastic anaemia was part of a hereditary syndrome due to selective malabsorption of vitamin B12 without intrinsic factor deficiency. Her 10 cases were later reinvestigated by Imerslund and Bjørnstad (1963), using the Schilling test, and they found that in all of them there was a very low urinary excretion of B12 which did not increase when intrinsic factor concentrate

was added. They also confirmed the finding of Colle, Greenberg, and Krivit (1961) that the gastric juice of such patients can act as a substitute for intrinsic factor in adult cases of pernicious anaemia. Other cases have been reported by Gräsbeck, Gordin, Kantero, and Kuhlback (1960), Lamy, Besançon, Loverdo, and Afifi (1961), Chavelet, Najean, Ravailleau, Grenet, and Bernard (1964), Sievens (1964), and Spurling, Sacks, and Jiji (1964). Some of them were sibs. The evidence thus obtained suggests that some other factor, possibly enzymatic in origin, is required in addition to intrinsic factor for the active absorption of B12, and that this factor is absent in these children. Spurling *et al.* (1964) regard it as a variety of juvenile pernicious anaemia which they propose should be subdivided into two types, one due to lack of intrinsic factor, and the other to selective malabsorption of B12. They point out that these two types have never been encountered in the same family, and that they can be distinguished quite simply by the presence of unexplained persistent proteinuria. It would, however, be less confusing if, in the present state of our knowledge, the term juvenile pernicious anaemia were restricted to those cases showing intrinsic factor deficiency.

The following case illustrates a further example of this rare variety of true pernicious anaemia in childhood; it also illustrates the delay in diagnosis which can occur if modern methods of investigation are not used.

Case Report

History. Valerie H. was born at full term on November 29, 1951 by caesarean section, birth weight 8 lb. 12 oz. (4 kg.). Her father was then 36 and her mother 25 years of age, with no history of consanguinity. There was one sister of 2 years. They were all in good health. The mother had a mild anaemia between the ages of 7 and 10 years and the paternal grandmother had been 'anaemic all her life'.

The child developed normally until the age of 1½ years, when she had an attack of 'dysentery' which lasted a week and left her listless, fretful, flabby, anorexic, and pale. As there was no improvement after 7 weeks on iron therapy, she was admitted to hospital where she was found to be underweight, at the 25th centile, and very

pale. Her tongue was normal, the liver and spleen were not felt, and there was no lymphadenopathy. Hb was 5.3 g./100 ml., red cells 1,280,000/c.mm., and the reticulocytes 0.4%. The white cells were 9,000/c.mm. Tests for occult blood were negative but her urine was mildly infected with coliform organisms and *B. proteus*. Although at the time the marrow was thought to be normoblastic, on reviewing the films there is undoubtedly some degree of megaloblastic change (Prof. Davidson). The fasting gastric juice contained no 'free acid' but had a total acidity of 25 mEq/l. The anaemia was treated with a total of 12 ml. of a parenteral liver preparation, 'Anahaemin'. 11 days after admission the blood picture remained unchanged, and one pint of fresh blood was given which raised her Hb from 4.7 g. to 14.3 g.

In October 1958 she was again examined following an attack of gastro-enteritis and her haemoglobin was found to be 11 g./100 ml. In June 1959 she was again admitted, at the age of 7½ years, after 3 weeks of abdominal pain and vomiting. She was very pale, breathless, easily fatigued, anorexic, and constipated. Her weight was still below the 25th centile, and there was no enlargement of the liver, spleen, or lymph nodes. The tongue and central nervous system were normal. Hb was 6.3 g./100 ml., red cells 1,900,000/c.mm., the mean corpuscular diameter 7.6 μ, white cells 3,000/c.mm., platelets 75,000/c.mm., and bone-marrow megaloblastic. The direct antiglobulin (Coombs) test was negative and red cell fragility was normal. A barium meal and follow-through was normal. Vitamin B12 100 μg. was given intramuscularly daily for 5 days together with 30 mg. folic acid orally, ascorbic acid, and ferrous gluconate. Despite this, her blood picture deteriorated, and after five days she was transfused one pint of packed red cells which raised her Hb from 5.7 g. to 12.3 g. She continued on folic acid and ascorbic acid for 2½ years. In January 1960, Hb was 13.6 g./100 ml. but by April it had fallen to 10.5 g., and she was given 5 intramuscular injections of an iron-dextran complex ('Imferon'). Her Hb rose to 11.5 g.

Present admission. Five days before her third admission on October 9, 1962 to Sydenham Children's Hospital, under the author's care, at the age of 11 years,

she became pale with abdominal pain, anorexia, and vomiting. She presented with a waxy pallor and an icteric tinge, but her tongue was still normal, and there was still no enlargement of the liver, spleen, or lymph nodes. Although no abnormality was detected in the central nervous system, she sometimes complained of 'pins and needles' in her feet at night. Her weight of 31.3 kg. was still below the 25th centile, and her height of 132 cm. was only at the third centile. She had hazel eyes and fair hair. The Hb was 5.4 g./100 ml., red cells 1,620,000/c.mm., reticulocytes 0.5%, white cells 2,500/c.mm., platelets 71,000, blood sedimentation rate 43 mm. in one hour (Westergren), serum bilirubin 1 mg./100 ml., direct antiglobulin test negative; red cell fragility, bone-age, intravenous pyelogram, urine, and stools were all normal. The bone-marrow was now frankly megaloblastic. A D-xylose absorption test was normal: 5 hours after a 25 g. dose the blood level reached 78 mg./100 ml. and the urine contained 10.7 g. of xylose. A four-day fat balance test showed a fat absorption of 99.1%. Free acid was present in the gastric juice, as demonstrated both by the tubeless ('Diagnex') test and by the augmented histamine test. The serum B12 was 55 μμg./ml., and the FIGLU test, after histidine loading, showed no increased urinary excretion. Intestinal and gastric biopsies (Dr. L. Fry) were reported on by Dr. R. M. H. McMinn as showing no villous atrophy and no histological or histochemical abnormality. The child was started on 20 μg. vitamin B12 daily, and three days later her reticulocytes had risen to 26%. After a month on B12, totalling 280 μg. of cyanocobalamin, her Hb reached 13.3 g./100 ml. and the platelets were 156,000/c.mm. On monthly injections of 100 μg. she has remained in complete remission up to the present time.

Dr. D. Doniach kindly investigated the sera from the patient, her sister, and her parents for auto-antibodies, and the results are shown in Table I. The only positive findings were in the mother and sister; the mother possessed antibodies for thyroid cytoplasmic antigen, and was weakly positive in the immunofluorescence test for antibodies to the second antigen of the thyroid acinar colloid. The sister showed a trace of thyroglobulin antibodies (tanned red cell agglutination titre 1/20 specifically inhibited by thyroglobulin).

TABLE
Autoantibody Production

Subject	Age (yr.)	Autoantibodies							Antinuclear Factors
		Gastric			Thyroid				
		Parietal Cells		Intrinsic Factor	Tanned Red Cell	Second Acinar Colloid	Cyto-plasmic	Comple-ment Fixation	
		Cyto-plasmic	Comple-ment Fixation						
Valerie	11	-	-	-	-	-	-	-	-
Father	48	-	-	-	-	-	-	-	-
Mother	37	-	-	-	-	±	+	-	-
Sister	13	-	-	-	±	-	-	-	-

± = weak.

Further investigations at the Experimental Haematology Research Unit at St. Mary's Hospital (Dr. S. Ardeman) showed that the patient had a serum vitamin B12 (*L. leishmanii*) level of 500 $\mu\text{g./ml.}$, and a serum folic acid (*L. casei*) level of 9.0 $\text{m}\mu\text{g./ml.}$, both normal. In a Schilling test she absorbed a subnormal amount of $^{58}\text{Co-B12}$, the urinary excretion being only 1.1% in 24 hours. Her gastric juice failed to promote absorption of an oral dose of radioactive vitamin B12 in a patient with proven pernicious anaemia (urinary excretion 4.6%) who subsequently absorbed normally (urinary excretion 15.2%) when given a similar dose of normal gastric juice. An *in vitro* assay was carried out on 23 ml. gastric juice collected in one hour following an augmented dose of histamine; free acid was plentiful (pH 1.5), but intrinsic factor concentration was very low (5 units/ml.) and her total secretion of intrinsic factor was only 100 units. This is in the pernicious anaemia range. Investigation of the parents gave the following results.

Mother. Routine blood tests normal; serum vitamin B12 265 $\mu\text{g./ml.}$ In the hour following an augmented dose of histamine she secreted 135 ml. of acid gastric juice (pH 1.2), but her total output of intrinsic factor was only 2,000 units. This is a low normal value and may represent some degree of intrinsic factor synthesis defect. Her Schilling test was normal (urinary excretion 12.5%).

Father. Routine blood tests normal; serum vitamin B12 300 $\mu\text{g./ml.}$ He secreted 115 ml. of gastric juice (pH 1.4) in the hour after an augmented dose of histamine and his total output of intrinsic factor during that time was 3,700 units. The Schilling test was vitiated by urine loss.

Discussion

The essential abnormality in pernicious anaemia is the failure of gastric secretion of an intrinsic factor needed for the absorption of the extrinsic factor vitamin B12. The present case appears to be one which can be accepted as an example of pernicious anaemia in childhood, from the virtual absence of intrinsic factor *in vivo* and *in vitro*, resulting in a chronic megaloblastic anaemia which relapsed when specific therapy was withheld but which remained corrected by parenteral vitamin B12. Malabsorption was ruled out by the demonstration of a normal fat and xylose excretion and a normal intestinal mucosa. An interesting feature of the case was that only 6 injections of liver extract given before the age of 2 years carried her through for nearly 6 years before she again became seriously anaemic. She then received enormous doses of folic acid (30 mg. daily over a period of 2½ years) which did no damage to her nervous system, in contrast to the neurological involvement described by Pearson *et al.* (1964) after smaller doses given over a much shorter time. 9 months after ceasing to take folic acid she once again

developed severe anaemia, an interval more in keeping with reports indicating that relapse after withholding specific therapy occurs usually within 6 to 18 months. Normal gastric secretion was then shown to be present, which is a common finding in juvenile pernicious anaemia, in contrast to the disease in adults, in whom lack of intrinsic factor secretion has been thought to be secondary to atrophy of the gastric mucosa with achlorhydria (Callender and Denborough, 1957). This difference between adult and childhood pernicious anaemia raises the question as to whether the disease is one and the same.

It is true that in both there is a strong familial tendency. In adults, Callender and Denborough (1957) found that 19% of their 142 families contained 2 or more cases, and they claimed they could detect an asymptomatic heterozygous carrier state in many relatives of pernicious anaemia patients. This was confirmed by McIntyre *et al.* (1959) who, by the extensive use of the Schilling test, showed that low absorption of B12 was present in up to 38% of relatives, suggesting that patients inherit a dominant autosomal gene responsible for the deficient secretion of intrinsic factor, and that they develop the disease when, according to Mollin, Baker, and Doniach (1955), atrophy of the gastric mucosa and achlorhydria eventually supervene to cause further deficiency of intrinsic factor. These findings are closely paralleled by autoimmune studies. Gastric parietal cell antibodies have been found in 89% of adults (Doniach and Roitt, 1964) and in 36% of their relatives (Doniach, Roitt, and Taylor, 1965), who, though asymptomatic, may nevertheless show a varying degree of chronic atrophic gastritis, achlorhydria, and deficient vitamin B12 absorption (Coghill, Doniach, Roitt, Mollin, and Williams, 1965). Thyroid antibodies have often been found as well (Doniach and Roitt, 1964), but intrinsic factor antibodies only if pernicious anaemia is present and even then only in 57% of cases (Ardeman and Chanarin, 1963). It has been suggested by Doniach and Roitt (1964) that pernicious anaemia in the absence of intrinsic factor antibodies may be due to a genetic failure of intrinsic factor synthesis, either alone or in combination with autoimmune disease. Nevertheless, it can be concluded that most cases of pernicious anaemia in adults are secondary to autoimmune gastritis determined by an inherited abnormality of the immunological mechanisms related to organ-specific antigens (Doniach *et al.*, 1965).

On the other hand, in children, autoimmune studies have been largely negative. In 9 of the cases which started in infancy (including the present

one), there was a complete absence of gastric and thyroid antibodies, whereas in 3 children who developed pernicious anaemia between the ages of 10 and 12 years there was gastric mucosal atrophy with achlorhydria, and though antibodies against intrinsic factor were demonstrated, there were none against parietal cells and thyroid (Doniach *et al.*, 1965). Furthermore all antibody tests were negative in our patient's family except for weak thyroid antibodies in the mother and sister. There is thus no evidence for assuming that pernicious anaemia commencing in early childhood is an autoimmune disease, but there may be grounds for believing that it is due to a congenital defect in intrinsic factor synthesis which is inherited, since, as with adults, there is a strong family history.

Among the 14 uncomplicated cases of pernicious anaemia in children hitherto reported there were no fewer than 4 pairs of sibs, with consanguinity of the parents only in the cases of Mollin *et al.* (1955) and Pearson *et al.* (1964), and a similar familial tendency is found even when the disease is complicated by hypoparathyroidism. Unfortunately few family studies have been carried out, and the Schilling test has been performed on both parents in only 5 instances. It is of interest that Waters and Murphy (1963) found that both parents and the unaffected sibs of their patients were heterozygotes, and they thought that homozygous inheritance of the gene could explain the almost complete suppression of intrinsic factor secretion in infancy and the appearance of the disease as soon as the stores of vitamin B₁₂, built up by placental transfer, had become exhausted. The cases of Mollin *et al.* (1955) and of Lambert *et al.* (1961) conformed to this view, but those of Leikin (1960) and Pearson *et al.* (1964) did not. Moreover, in the present case the mother alone is a possible heterozygote since, though her serum vitamin B₁₂ and Schilling test were normal, an *in vitro* assay showed defective intrinsic factor production, possibly due to an enzyme defect. In contrast the father secreted intrinsic factor normally.

The mode of inheritance must, therefore, remain obscure until more cases are reported with family studies. The quantitative measurement of intrinsic factor, according to the method of Ardeman and Chanarin (1965), may be able to provide more information in this regard than can the Schilling test alone. Further work will be needed before deciding that childhood and adult pernicious anaemia arise by different mechanisms, but the scanty evidence so far available is pointing in this direction.

Summary

A girl, now 11 years old, is described, who has

been shown to be suffering from pernicious anaemia which produced symptoms first when she was under the age of 2.

A congenital genetically determined defect in intrinsic factor synthesis is considered to be the likely cause, in contrast to the genetically determined disorder of the immunological mechanisms, which characterizes most of the cases of pernicious anaemia in adults.

It is suggested that the quantitative measurement of the intrinsic factor in close relatives may help to elucidate the mode of inheritance of juvenile pernicious anaemia.

I am very grateful to Prof. W. M. Davidson and to Dr. D. Doniach for their great interest and help with this case and for their valuable comments on the manuscript, to Drs. L. Fry and R. M. H. McMinn for their work in relation to the biopsies, to Dr. S. Ardeman for carrying out the vital radioactive tests and quantitative intrinsic factor assays, and to Dr. J. Keall for the laboratory investigations.

REFERENCES

- Ardeman, S., and Chanarin, I. (1963). A method for the assay of human gastric intrinsic factor and for the detection and titration of antibodies against intrinsic factor. *Lancet*, **2**, 1350.
- , and — (1965). Assay of gastric intrinsic factor in the diagnosis of Addisonian pernicious anaemia. *Brit. J. Haemat.*, **11**, 305.
- Callender, S. T., and Denborough, M. A. (1957). A family study of pernicious anaemia. *ibid.*, **3**, 88.
- Chavelet, F., Najean, Y., Ravailleau, J., Grenet, P., and Bernard, J. (1964). Étude d'un cas d'anémie pernicieuse de l'enfant avec activité 'facteur intrinsèque' normale du suc gastrique. *Nouv. Rev. franç. Hémat.*, **4**, 311.
- Clement, D. H., Nichol, C. A., and Welch, A. D. (1961). A case of juvenile pernicious anemia; study of the effects of folic acid and vitamin B₁₂. *Blood*, **17**, 618.
- Coghill, N. F., Doniach, D., Roitt, I. M., Mollin, D. L., and Williams, A. W. (1965). Autoantibodies in simple atrophic gastritis. *Gut*, **6**, 48.
- Colle, E., Greenberg, L., and Krivit, W. (1961). Studies of a patient with selective deficiency in absorption of vitamin B₁₂. *Blood*, **18**, 48.
- Donegan, W. L., and Spiro, H. M. (1960). Parathyroids and gastric secretion. *Gastroenterology*, **38**, 750.
- Doniach, D., and Roitt, I. M. (1964). An evaluation of gastric and thyroid auto-immunity in relation to haematologic disorders. *Seminars Hemat.*, **1**, 313.
- , —, and Taylor, K. B. (1965). Autoimmunity in pernicious anemia and thyroiditis: a family study. *Ann. N.Y. Acad. Sci.*, **124**, 605.
- Gräsbeck, R., Gordin, R., Kantero, I., and Kuhlback, B. (1960). Selective vitamin B₁₂ malabsorption and proteinuria in young people: a syndrome. *Acta med. scand.*, **167**, 289.
- Hung, W., Migeon, C. J., and Parrott, R. H. (1963). A possible autoimmune basis for Addison's disease in three siblings, one with idiopathic hypoparathyroidism, pernicious anemia, and superficial moniliasis. *New Engl. J. Med.*, **269**, 658.
- Imerslund, O. (1960). Idiopathic chronic megaloblastic anemia in children. *Acta paediat. (Uppsala)*, **49**, Suppl. 119.
- , and Bjørnstad, P. (1963). Familial vitamin B₁₂ malabsorption. *Acta haemat. (Basel)*, **30**, 1.
- Lambert, H. P., Pranker, T. A. J., and Smellie, J. M. (1961). Pernicious anaemia in childhood. *Quart. J. Med.*, **30**, 71.
- Lamy, M., Besançon, F., Loverdo, A., and Afifi, F. (1961). La malabsorption spécifique de la vitamine B₁₂ avec protéinurie. *Arch. franç. Pédiat.*, **18**, 1109.

- Leikin, S. L. (1960). Pernicious anemia in childhood. *Pediatrics*, **25**, 91.
- McIntyre, P. A., Hahn, R., Conley, C. L., and Glass, B. (1959). Genetic factors in predisposition to pernicious anemia. *Bull. Johns Hopk. Hosp.*, **104**, 309.
- Mollin, D. L., Baker, S. J., and Doniach, I. (1955). Addisonian pernicious anaemia without gastric atrophy in a young man. *Brit. J. Haemat.*, **1**, 278.
- Morse, W. I., Cochrane, W. A., and Landrigan, P. L. (1961). Familial hypoparathyroidism with pernicious anemia, steatorrhea, and adrenocortical insufficiency. *New Engl. J. Med.*, **264**, 1021.
- Pearson, H. A., Vinson, R., and Smith, R. T. (1964). Pernicious anemia with neurologic involvement in childhood. *J. Pediat.*, **65**, 334.
- Reisner, D. J., and Ellsworth, R. M. (1955). Coexistent idiopathic hypoparathyroidism and pernicious anemia in a young girl. *Ann. intern. Med.*, **43**, 1116.
- Sievens, C. J. (1964). Megaloblastic anemia in a 3-year-old child. *Blood*, **23**, 547.
- Spurling, C. L., Sacks, M. S., and Jiji, R. M. (1964). Juvenile pernicious anemia. *New Engl. J. Med.*, **271**, 995.
- Waters, A. H., and Murphy, M. E. B. (1963). Familial juvenile pernicious anaemia; a study of the hereditary basis of pernicious anaemia. *Brit. J. Haemat.*, **9**, 1.
- Wilkins, L. (1957). *The Diagnosis and Treatment of Endocrine Disorders in Childhood and Adolescence*, 2nd ed., p. 430. Thomas, Springfield, Illinois.

Addendum

McIntyre, Sullivan, Jeffries, and Silver (1965) have reported another case of pernicious anaemia starting in infancy; they refer to 4 others, including one of late childhood arising in association with endocrine disease, that were investigated by Herbert, Streiff, and Sullivan (1964) but few clinical details are given. Other infantile cases inadvertently omitted from the text of this paper include one by Oehme, Hundeshagen, and Willenbockel (1962) and another by Moody (1965), but both were incompletely investigated.

REFERENCES

- Herbert, V., Streiff, R. R., and Sullivan, L. W. (1964). Notes on vitamin B₁₂ absorption; autoimmunity and childhood pernicious anemia; relation of intrinsic factor to blood group substance. *Medicine (Baltimore)*, **43**, 679.
- McIntyre, O. R., Sullivan, L. W., Jeffries, G. H., and Silver, R. H. (1965). Pernicious anemia in childhood. *New Engl. J. Med.*, **272**, 981.
- Moody, E. A. (1965). Juvenile pernicious anemia. *Northw. Med. (Seattle)*, **64**, 981.
- Oehme, J., Hundeshagen, H., and Willenbockel, U. (1962). Über die perniziöse anämie des Kindes. Zugleich ein Beitrag zur brauchbarkeit des urinexkretionstestes nach Schilling. *Z. Kinderheilk.*, **87**, 289.