temperature and low bicarbonate content of the medium for optimal growth in tissue culture. Twenty-four strains grew only in human-kidney cultures and resembled F.E.B. virus of Salisbury. Seven viruses grew in both human-kidney and monkey-kidney cultures and resembled but were not identical to H.G.P. virus of Salisbury. E.C.H.O. 28 virus, as well as these new viruses, was associated with an outbreak of colds in October, 1960. Another outbreak in late January and early February was caused primarily by the respiratory syncytial virus which was isolated from 11 specimens. Para-influenza viruses 1 and 2 appeared sporadically.

Only one virus was isolated from 191 specimens obtained during convalescence. A similar agent had been isolated from the acute specimen of the same student. The 456 specimens obtained from asymptomatic individuals throughout the study period yielded only two viruses, both growing only in human-kidney cultures.

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On April 16 next year regulations relating to the lead content of food, based on recommendations of the Food Standards Committee, will come into operation in England and Wales. The regulations lay down statutory limits for the lead content of food and beverages imported, sold, or intended for sale for human consumption. The regulations limit, with certain exceptions, the lead content of ready-to-drink non-alcoholic beverages to 0.2 parts per million and of foods to 2.0 p.p.m. One of the exceptions is fish (including crustacea and molluscs or any product containing such fish) for which a natural lead content in excess of 2.0 p.p.m. has been established. The Secretary of State for Scotland is making corresponding regulations to apply in Scotland, and similar regulations will also be made for Northern Ireland. (H.M.S.O., price 5d. net.)

HYPOPLASTIC ANAEMIA TREATED BY TRANSFUSION OF FOETAL HAEMOPOIETIC CELLS

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Aplastic anaemia is an uncommon condition arising from many causes, but most frequently idiopathic. Both course and prognosis are extremely variable. Spontaneous remission has often been recorded, so that any benefit which follows a particular type of treatment must be cautiously interpreted. In 1958 Dameshek, McFarland, and Granville (1960) reported some remissions after bone-marrow transplantation, and it therefore seems pertinent to place on record the results of similar treatment with foetal haemopoietic cells.

Fourteen cases have received foetal liver cells by intravenous injection and have survived more than 48 hours after their administration (see Table). From this series two cases are presented in detail.

Case 9

A white male born in 1923 began to notice symptoms attributable to anaemia in 1954. This was confirmed in September, 1956. Subsequently he required the transfusion of more than 3 pints (1.7 litres) of blood each month. The usual haematinics and cortisone (300 mg. daily initially; a six-weeks course) conferred no benefit. He came of a healthy stock and had not been in contact with known myelotoxic agents.

Examination in July, 1958, revealed a pallid, normally developed man without evidence of a haemorrhagic diathesis or other abnormal physical signs.

Investigations.—Sedimentation rate 50 mm. in first hour (Westergren). Urine analysis normal. Haemoglobin, 7.4 g./100 ml.; M.C.H.C., 31%; M.C.V., 108 cubic microns; reticulocytes, 0.2–0.4%; total leucocyte count, 2,400/c.mm. (neutrophils 528, lymphocytes 824); platelets, 49,000/c.mm. Group O rhesus-positive. Direct antiglobulin test negative. An aspiration biopsy of the marrow showed marked hypoplasia involving all myeloid elements, a finding confirmed by iliac-crest-trephine biopsy (Fig. 1). The red-cell half-life (^{51}Cr method) was slightly reduced (19 days; normal 22–30 days). The half-time of the disappearance of iron from the serum re-utilization was reduced to 40% (normal 90–100%) in 10 days (^{59}Fe method). The Ham and Donath-Landsteiner tests were negative. Erythrocyte osmotic fragility was normal (fresh and incubated). Serum vitamin B₁₂ was 700 $\mu\text{g.}/\text{ml.}$ Liver-function tests were normal, including the serum albumin (5.3 g.), globulin (1.8 g.), and electrophoresis. The Wassermann reaction was negative and the blood urea 35 mg./100 ml. The serum was fully saturated with iron at 245 $\mu\text{g.}/100\text{ ml.}$ A histamine-fast achlorhydria was present. The stools were negative for occult blood. The 24-hour urinary excretion of 17-keto-steroids was 8.4 mg. Radiographs of the chest and skeleton

were normal. A diagnosis of idiopathic hypoplastic pancytopenia was made.

Foetal liver (about 3×10^9 cells) was injected intravenously in August, 1958, with cortisone cover. Within the next few days the reticulocytes rose, but only to 2%. Transfusion requirements in the following year were 19 pints (10.8 litres), compared with approximately 35 pints (19.9 litres) in the

FIG. 1.—Case 9. Trephine specimen from iliac crest. ($\times 15.5$.)

FIG. 2.—Case 9. Chart showing rate of decline of haemoglobin in relation to treatment.

previous year. However, the transfusion requirements may have been decreasing during the early part of 1958—that is, before the implant was carried out (see Fig. 2).

He was readmitted in September, 1959, when the haemoglobin was 9.5 g./100 ml., reticulocytes 0.4%, total leucocytes 3,300, and platelets 32,000/c.mm. The marrow picture was unchanged. Three further intravenous injections of foetal liver (about 14×10^9 cells) were given at the end of that month without corticosteroid cover. The reticulocytes rose to 8% and the platelets to 81,000/c.mm. five days later. When a sample of this blood was centrifuged by the method of Constandoulakis and Kay (1959) a reticulocyte-rich layer (70–80% reticulocytes) was obtained which was tested serologically for the presence of donor cells, but these could not be detected. The last transfusion was given in December, 1959. Since then the haemoglobin has risen spontaneously to 14.2 g./100 ml. The transfusion requirements were:

December, 1956–December, 1957, about 38 pints (21.6 litres).

December, 1957–December, 1958, 21 pints (11.9 l.) (transplant August, 1958).

December, 1958–December, 1959, 20 pints (11.4 l.) (transplant September, 1959).

December, 1959–December, 1960, 4 pints (2.3 l.) (December, 1959).

It was not possible to establish the genotype of this patient before injection of foetal cells. In August, 1960, he was found to be: O, cDe/cde, N, S⁺, P₁+++⁺, Lu^a–, K–, Le^a–, Le^b+, Fy^a+. There was no evidence of mixed erythrocyte populations. His parents are: O, cDe/cde, MN, S⁺, P₁+++⁺, Lu^a+, K–, Le^a+, Le^b–, Fy^a–; O, cDe/cde, MN, S–, P₁+++⁺, Lu^a–, K–, Le^a–, Le^b+, Fy^a+. The genotypes of the foetal donors were:—August, 1958: O, CDe/cde, MN, S⁺, K–, Fy^a+. September, 1959: (6 $\times 10^9$) O, cde/cde, MN, S⁺, P₁–, Lu^a–, K–, Fy^a–; (6 $\times 10^9$) O, cDe/cde, M, S⁺, P₁–, Lu^a–, K–, Fy^a–; (2 $\times 10^9$) O, CDe/CDe, N, S–, P₁+, Lu^a–, K–, Fy^a–.

Comment.—This may well have been a slow but spontaneous recovery, but certain points are of interest. Firstly, for four weeks after the initial injection of foetal cells the haemoglobin not only failed to fall but actually rose slightly. Although few reticulocytes were in the circulation a number of normoblasts could be detected in the first two weeks of this time. Early in 1959 the question arose whether to repeat the injection of foetal cells in consequence of the slight and questionable benefit derived from the first injection. It was decided to wait and record the rate of haemoglobin decline for a few more months and if this showed no further improvement or even regressed then a second treatment should be tried. In September, 1959, it was judged that these conditions were fulfilled (see Fig. 2), and the second treatment, using a larger dose of foetal cells, was given. After this only one further transfusion was given, and it is possible that even this was not necessary.

Case 6

A single white male (born in 1936) began to suffer from recurrent epistaxes in June, 1956. At that time he was serving with the Army in Malaya. In November he was found to be profoundly anaemic and thereafter required the transfusion of 2 to 3 pints (1.1 to 1.7 litres) of blood each month. There was little response to cortisone, at a dose of 300 mg. daily initially and 75–100 mg. daily as maintenance therapy, over a period of five months. No beneficial effect followed the usual haematinics or cobalt chloride (100 mg. daily for one month).

He came of healthy stock and had not been exposed to known myelotoxic agents. In October, 1958, examination revealed a pallid, normally developed young man without evidence of a haemorrhagic diathesis or other abnormal physical signs.

Investigations.—Sedimentation rate, 35 mm. in first hour (Westergren). Urine analysis, normal. Haemoglobin, 4.1 g./100 ml.; M.C.H.C., 31%; M.C.V., 93 cubic microns; reticulocytes, 0.1–1%; total leucocyte count, 1,750/c.mm.

FIG. 3.—Case 6. Trephine specimen from iliac crest. ($\times 15.5$.)

(neutrophils 560, lymphocytes 995); platelets, 15,000/c.mm. Group O rhesus-positive. Direct antiglobulin test negative. The cellularity of an aspirated specimen of marrow was greatly reduced. All elements were involved. This finding was confirmed by an iliac-crest-trephine biopsy (Fig. 3). The red-cell half-life (^{51}Cr method) was slightly reduced (20 days). The half-time of the disappearance of iron from the serum was prolonged (four to five hours), and the maximum re-utilization was reduced to 28% in eight days (^{59}Fe method). The serum was fully saturated with iron at 228 $\mu\text{g.}/100\text{ ml.}$ The serum albumin was 5.1 g. and globulin 1.2 g./100 ml. Electrophoresis was normal. The stools did not contain occult blood. Faecal urobilinogen per 24 hours was 47, 60, and 45 mg. The Wassermann reaction was negative. The blood urea was 35 mg./100 ml. The gastric juice contained free acid. Radiographs of the chest and skeleton were normal. A diagnosis of idiopathic hypoplastic pancytopenia was made.

At the end of October, 1958, he received four intravenous injections of foetal liver (about 14×10^9 cells) with a cortisone cover (see Fig. 4). No reticulocyte response was

FIG. 4.—Case 6. Chart showing rate of decline of haemoglobin in relation to treatment.

observed, but six days later the platelet count rose to 77,000/c.mm. and remained at this level subsequently. Simultaneously there was some increase of leucocytes from around 2,000 to about 4,000/c.mm. and this was also maintained. Prednisolone was continued at a dose of 10 mg. daily. The transfusion requirements were:

November, 1956–November, 1957, about 27 pints (15.3 l.).

November, 1957–November, 1958, 28 pints (15.9 l.) (transplant October, 1958).

November, 1958–November, 1959, 21 pints (11.9 l.) (last transfusion September, 1959).

November, 1959–November, 1960, no transfusions.

Since the last transfusion in September, 1959, the haemoglobin level has been maintained in the region of

10 g./100 ml. There has probably been a slight fall since, but the patient remains well and in full employment. A further injection is being considered.

Comment.—The lack of data from which the rate of haemoglobin decline could be estimated at each stage of the disease makes it difficult to assess the possible effect of treatment in this case. The probability is that recovery was spontaneous and unrelated to the foetal liver injections.

In the other 12 cases (see Table) there was no demonstrable long-term benefit, although in some who received fresh cells the appearance of normoblasts in the peripheral circulation a few days after injection probably indicated a temporary survival of the grafted cells (for details see Constandoulakis, 1959). In all cases the cells were prepared and administered by methods which have already been described (Kay and Constandoulakis, 1959, 1960), and it is worth noting that with one exception, where rapid injection caused some nausea and vomiting, no untoward side-effects occurred. Furthermore, in those cases where death occurred within a few days of injection no evidence of pulmonary embolism was observed at necropsy.

Discussion

The 14 cases treated represent a wide variety of forms of marrow aplasia. The term is used here in a comprehensive sense to include all types of marrow hypofunction. Two patients in whom an ultimate diagnosis of leukaemia was made could perhaps be excluded, although at the time of treatment their condition more closely resembled aplasia. The four children treated included two with a familial history, and all were different with regard to the type of aplasia present.

Of the eight remaining adult cases, four were roughly speaking acute, with pancytopenia, and four were chronic, presenting with anaemia but with deficiency of granulocytes and platelets also. These latter include the two in which remission has occurred. The patients form a selected series in that the acute cases were referred for "marrow" therapy when other methods had failed. The chronic cases had also been treated in other ways, including steroids in three; but these four patients were probably fairly typical of the prolonged variety of marrow hypoplasia.

Even this subdivision of the disease is known to pursue a very variable course with a fairly high rate of spontaneous remission. Exactly what this rate amounts

Details of Cases

No.	Age	Sex	Type of Disease	Foetal Liver Cells	Response
1	18 months	M	Erythrogenesis imperfecta	Fresh cells (19.5×10^9)	No response
2	9 years	M	Neutropenia (? familial)	Stored cells only (2.4×10^9)	" " Early death
3	9 "	M	Hypoplasia, mainly erythropenia	Fresh cells (9.5×10^9)	Temporary normoblastaemia. No long-term improvement
4	9 "	F	Hypoplasia, mainly thrombocytopenia	Stored cells only ($c. 20 \times 10^9$)	No response. Early death
5	17 "	M	Acute pancytopenia	" " (20×10^9)	" " "
6	21 "	M	Chronic "	Fresh and stored cells (5.5 and 8.7×10^9)	Partial remission later (see text)
7	24 "	M	" "	Fresh cells (6×10^9)	Normoblastaemia with 10% foetal haemoglobin. No long-term response. Died 6 months later
8	29 "	M	Acute "	Fresh and stored cells ($c. 7$ and 8×10^9)	Progressive deterioration and death
9	34 "	M	Chronic "	Fresh cells ($c. 17 \times 10^9$)	Remission (see text)
10	49 "	M	Acute "	Fresh and stored cells (4.1 and 3.3×10^9)	No response. Early death
			(? chloramphenicol)		
11	54 "	M	Acute pancytopenia	Stored cells only (15×10^9)	" " " " (cellular marrow at necropsy)
12	55 "	F	Aplasia progressing to leukaemia	Fresh and stored cells (3.5 and 7.5×10^9)	Temporary normoblastaemia with primitive myeloid cells progressing to type of chronic leukaemia
13	58 "	F	Chronic pancytopenia	Fresh and stored cells ($c. 16$ and 4.5×10^9)	No response. Early death
14	67 "	F	Aplasia progressing to leukaemia	Fresh cells (2×10^9)	Transformed to blast-cell leukaemia

to cannot be estimated. In the absence of exact diagnostic criteria, cases will vary from series to series, while those that get published may well include an abnormally high proportion of successes. Again, some cases appear to be benefited by steroid therapy, others by splenectomy. Since remission is usually a slow process it is often impossible to distinguish the results of therapy from the natural fluctuations of the disease. As Scott, Cartwright, and Wintrobe (1959) point out, even the excellent results attributed to splenectomy may well be largely accounted for by selection of the more chronic cases, which are inherently more likely to recover spontaneously.

In the light of these considerations it is impossible to assess the true rate of spontaneous remission at all accurately. The interested reader must perforce read in detail the reviews of Boon and Walton (1951), Heaton, Crosby, and Cohen (1957), Scott *et al.* (1959), and Israëls and Wilkinson (1961). It is not unlikely, however, that somewhat less than one-third of such cases as are commonly labelled aplastic anaemia may recover spontaneously.

Clearly the cases here reported represent no particular advance, and no firm conclusions can be reached. At the same time it is worth noting that in Case 9 the administration of a small dose of foetal haemopoietic cells was followed by lengthening of the intervals between transfusions, and that when the rate of haemoglobin decline had again accelerated, a second, larger, dose of foetal cells was followed by a more complete remission. It is not impossible that in this case, as in those reported by Boon and Walton (1951), Morrison and Samwick (1940), Dameshek, McFarland, and Granville (1960), and McFarland, Granville, Schwartz, Oliner, Misra, and Dameshek (1961), a temporary proliferation of foreign metabolically competent cells in the marrow is able to help the impaired cells to recover, or that a direct humoral factor is involved.

In this context the experimental work of Bernstein and Russell (1959) should be cited. They have shown that genetically anaemic mice may, without prior irradiation, be cured by transplantation of foetal liver cells from closely related animals. It can be proved that the donors are not quite isologous, since skin grafts are slowly rejected. The probability here is that the foreign cells are able to withstand a weak homograft reaction and so colonize the recipient marrow, although another mechanism such as transduction cannot be entirely excluded.

Meanwhile Diamond (1960) has revealed the therapeutic potency of androgens in many cases of aplasia, and it seems likely that this will take first place in future therapeutic efforts.

Summary

Fourteen cases representing different types of bone-marrow hypoplasia have been treated by injection of foetal haemopoietic cells. Out of four cases of chronic pancytopenia two have had remissions, the timing of which suggests that the foetal cells may possibly have been partly responsible, although chimerism was not detectable.

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SYNCHRONOUS COMBINED ABDOMINO-PERINEAL EXCISION OF RECTUM

BY

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In the course of the evolution of any successful operative procedure certain steps can be traced: firstly, the tentative trials in the hands of the originator; secondly, the production of successful results from the departments of the specialists; and, finally, confirmation from the units of the general surgeon.

Since the synchronous combined abdomino-perineal excision of the rectum was first described by Lloyd Davies in 1939, the first two steps have been most adequately negotiated. Despite the increasing popularity of the operation there is little evidence available to indicate what results the average general surgical unit can anticipate. The purpose of this paper is to present the experiences of such a unit over the past 10 years.

Scope of the Survey

Over this period of 10 years 160 cases of carcinoma in the lower 10–12 cm. of the rectum have been admitted to one surgical unit at the Royal Infirmary, Sheffield. Lesions above this level have not been included in the series, as it has been our practice to submit these to sphincter-preserving operations. Of these 160 cases, 143 (89.4%) have undergone the operation of synchronous combined abdomino-perineal excision of the rectum. Of the remaining 17 cases (10.6%), four were judged to be inoperable because of very extensive local disease at the time of the initial examination or after an examination under anaesthesia. Nine others were submitted to laparotomy, but the operation was abandoned for a variety of reasons, the chief of these being extensive local spread with fixation laterally to the vascular pedicles or posteriorly to the sacrum; this occurred in six cases. Scattered metastases in an otherwise normal liver were not regarded as a contra-indication to excision of the rectum, but in three cases the hepatic secondaries were so gross that excision was