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THE PHYSIOLOGICAL MACROCYTOSIS OF PREGNANCY

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Summary

Sixty-four healthy women were followed during normal pregnancies in which they took supplements of iron *or* iron+folate *or* an inactive placebo. The red cell size increased in those patients receiving iron alone *or* iron+folate. The group on the inactive placebo who received no supplemental iron had red cells of constant size and a falling serum iron. Nineteen women not included in the trial developed macrocytosis during pregnancy: marrow samples from these showed normoblastic erythropoiesis in 18 and minor megaloblastic changes in one. Macrocytosis developing during pregnancy is a physiological change in the majority of cases.

THE normal newborn infant has larger red cells than the healthy adult. Macrocytosis may also be a consequence of disease: it is found in megaloblastic anaemia and in a number of pathological conditions where haemopoiesis remains normoblastic such as chronic alcoholism (Wu *et al*, 1974), myxoedema (Morton *et al*, 1976), marrow aplasia, haemolytic anaemia, sideroblastic anaemia and others. Many pregnant women have large red cells, and this study was undertaken to determine whether or not the macrocytosis in pregnancy is physiological or is pathological, perhaps due to deficiency of folate.

METHODS

The patients were 64 healthy white women with uncomplicated pregnancies. Primigravidae predominated, only 11 patients being in their second or a later pregnancy; 58 patients were between 20 and 30 years, only one patient in the series being under 20, and 5 over 30. The social classification followed the general pattern of the Maternity Unit Bookings in the hospital: 14 patients were in Class I, 18 in Class II, 25 in Class III, 6 in Class IV, none in Class V and one was unclassified. None of them had bacteriuria and only three patients smoked.

The project was approved by the Hospital Ethical Committee and all the women who were investigated agreed to take part after full explanation at their first visit to the antenatal clinic. This took place between 8th and 12th week of pregnancy. Blood samples were obtained at that time for routine investigations. The patients were asked to take no iron or folic acid preparations in the next month. When they returned 4 weeks after the initial visit they started one of three types of oral preparations which continued throughout the pregnancy. The subjects were allocated randomly and double blind to one of the three dosage schemes: ferrous fumarate supplying 100 mg of elemental iron *or* ferrous fumarate with 350 μg of folic

TABLE I

Mean haemoglobin, mean corpuscular volume, serum iron and red cell folate in 3 groups of pregnant women given a placebo, iron or iron+folate throughout pregnancy

Weeks pregnant	Haemoglobin (g/dl)			Mean corpuscular volume (MCV) (fl)			Serum iron (μmol/l)			Red cell folate (ng/ml)		
	Placebo (n = 13)	Fe (n = 35)	Fe+ Folate (n = 16)	Placebo	Fe	Fe+ Folate	Placebo	Fe	Fe+ Folate	Placebo	Fe	Fe+ Folate
First visit	12·4	12·6	12·4	86	86	85	–	–	–	–	–	–
12	12·1	12·5	–	87	86	84	19·0	18·9	16·9	265	317	297
16	12·2	12·2	12·1	85	86	86	18·3	17·2	17·9	292	318	304
20	11·9	12·0	11·8	87	87	86	16·9	17·4	17·0	259	327	293
24	11·8	12·0	11·7	87	88	87	16·0	19·6	19·6	267	302	365
28	11·8	12·2	11·8	87	89	88	14·6	20·5	19·1	270	276	412
32	11·5	12·3	12·2	87	89	87	14·3	21·6	19·7	264	301	451
36	11·8	12·8	12·4	86	88	87	11·1	24·4	21·4	304	288	468
40	11·2	12·8	12·3	86	90	88	–	20·7	17·2	–	288	400
6 weeks postnatal	12·8	13·4	12·8	85	86	85	12·3	17·2	16·7	239	252	396

acid, *or* a placebo containing neither iron nor folic acid. All of the preparations were packaged in identical sealed foil 'pop-up' containers, one tablet to be taken daily. The code was broken after a total of 64 patients had been followed. As the results appeared to be clear cut at this stage the study was terminated when 35 women had taken iron alone, 16 iron+folate and 13 the placebo. This distribution of numbers occurred entirely by chance.

All of the women were seen by one of us (I.M.) at a special antenatal clinic. The empty containers were collected every 4 weeks and a fresh supply of tablets given to the patient. A blood sample was taken at the start of therapy and every 4 weeks thereafter for measurement of the haemoglobin, red cell count, haematocrit and mean corpuscular volume (MCV) measured on the Coulter Counter Model S. The serum iron and iron binding capacity was also estimated in the specimen, as was the red cell folate (*Lactobacillus casei* assay). A further sample of blood was obtained 6 weeks after delivery. All the haemolysates for the red cell folate estimation (1 part whole blood, 9 parts 1 per cent ascorbate solution) were stored at −20 °C until the final specimen was obtained, when all the samples from a single patient were assayed together in a single batch.

An additional group of 19 women developed macrocytosis during pregnancy. Marrow aspirations were carried out on all of these.

Results

Haemoglobin, red cell count and haematocrit

The mean haemoglobin concentration in the iron group was 12·6 g/dl at the first visit and 12·8 g/dl at term; in the iron+folate group the concentrations were 12·4 and 12·3 g/dl. By contrast, the mean haemoglobin in the placebo group fell from 12·4 g/dl at the first visit to 11·2 g/dl at term (Table I). The red cell count and haematocrit values matched the haemoglobin values.

Mean corpuscular volume (MCV)

At the first visit the mean MCV was 85 to 86 fl in all three groups. In the group which received iron only the mean MCV rose to 90 fl at term and returned to 86 fl 6 weeks after delivery (Fig. 1). Those women who received folic acid as well as iron showed a similar rise from 85 fl at the first visit to 88 fl at term (Figs. 2 and 3). In the placebo group, however, the MCV did not change significantly

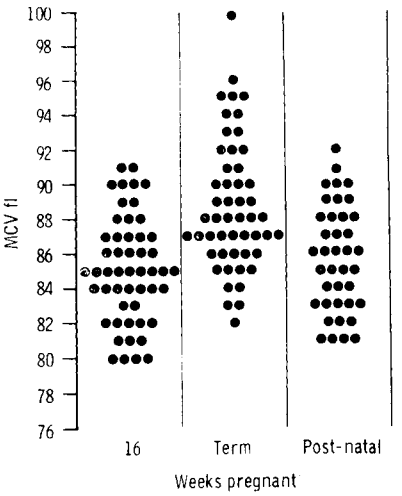

FIG. 1
Red cell mean corpuscular volume in women who received iron or iron+folate supplements.

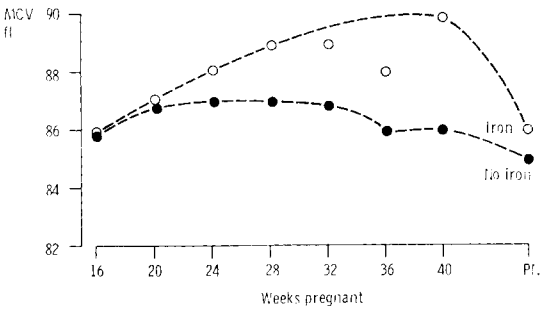

FIG. 2
Red cell mean corpuscular volume in women who received iron supplements or no iron.

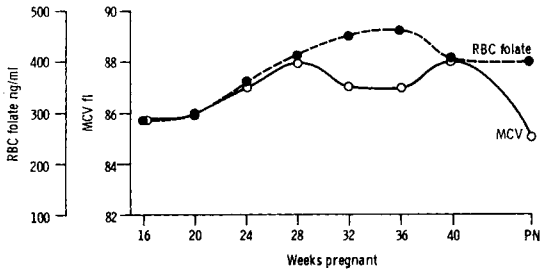

FIG. 3
Red cell folate and mean corpuscular volume in women given iron+folate supplements.

during the pregnancy; it was 86 fl both at the first visit and at term.

Iron status

The mean serum iron level was maintained in the two groups receiving oral iron, being at 12 weeks 16·9 μmol/litre and 18·9 μmol/litre respectively in the iron plus folate and iron groups, and at term 17·2 and 20·7 μmol/litre. In both groups there was a rise in the iron-binding capacity from 60·7 and 62·8 to 99·0 and 85·3 μmol/litre respectively. By contrast, the mean serum iron level in the placebo group fell from 19·0 μmol/litre at 12 weeks to 11·1 μmol/litre at 36 weeks while the iron-binding capacity showed a rise similar to the other groups.

Folate status

There was little change in the red cell folate values in the iron only and placebo groups, the values being 317 and 265 ng/ml at 12 weeks and 288 and 304 ng/ml at 36 weeks respectively. There were insufficient samples in the placebo group at term for analysis. In the group given the folic acid supplement the red cell folate increased from 297 ng/ml at 12 weeks to 468 ng/ml at 36 weeks and 400 ng/ml at term.

Statistical analysis

A linear regression of MCV on number of weeks pregnant was calculated for each patient, the 'slope' of the regression line being the rate of change of MCV (fl per week). The slopes of the placebo group were lower than the iron group ($P<0·001$) and also lower than the iron+folate group ($P<0·05$). The two treatment groups had slopes which were not significantly different ($P<0·05$).

The groups given iron *or* iron+folate also differed significantly from the placebo group in the slopes for the haemoglobin levels, haematocrit, mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), serum iron and red cell folate: the values of P ranged from 0·01 to 0·001. There was no significant difference between the placebo and other groups in red cell counts or in the rise of iron-binding capacity.

Marrow aspirations

While this survey was taking place, 19 women who were not included in the trial were investigated because their MCV had increased from a normal resting value early in pregnancy to 96–102 fl in the last trimester. Marrow samples were obtained from all of these women. Eighteen women showed normoblastic haemopoiesis and only one showed minor changes of megaloblastic type.

Discussion

Our data indicate that red blood cells increase in size in normal pregnancy provided that the patient is not iron deficient. This increase in MCV occurred in the group given folate supplements (and in whom the red cell folate levels increased in the expected manner) so it is not related to folate deficiency. Since the group given supplements of iron only showed the same rise in MCV as those with iron+folate it would appear that the abnormality lies in those who showed no change in red cell size but in whom the falling serum iron indicated the development of iron deficiency. Frank iron deficiency leads to a reduction of cell size with a marked decline in the MCV. Taylor and Lind (1976) interpreted a rise in MCV in an iron-supplemented group as a change induced by iron. We do not accept this interpretation and regard the failure of an iron-deficient group to show a rise in MCV as an abnormality and not as a physiological feature of pregnancy.

Our observations emphasize further the difficulty in making a diagnosis of megaloblastic anaemia in pregnancy in the UK. When the changes are well developed the blood findings offer no difficulty, but the early case is difficult to detect. Macrocytosis which is the most sensitive index of a megaloblastic process, for example in pernicious anaemia, occurs normally in pregnancy. White cell changes such as hypersegmented neutrophils, if clearly increased, are more helpful but these may be absent. They also occur in iron deficiency. Ancillary tests which measure folate status such as serum and red cell folate are also of limited value. The serum folate may be low in healthy pregnant women not taking folate supplements.

The red cell folate is still within the normal range in half the women in pregnancy who have

megaloblastic marrow changes (Chanarin, 1969). The deoxyuridine suppression test is almost always abnormal in megaloblastic anaemia but can give false positives in iron deficient pregnant women, although underlying folate deficiency cannot be excluded in these subjects (Taguchi and Chanarin, 1977).

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REFERENCES

Chanarin, I. (1969): *The Megaloblastic Anaemias.* Blackwell, Oxford.

Morton, L., Coburn, R. J., England, J. M., and Himsworth, R. L. (1976): *Quarterly Journal of Medicine*, **45**, 101.

Taguchi, H., and Chanarin, I. (1977): *British Journal of Haematology*, in press.

Taylor, D. H., and Lind, T. (1976): *British Journal of Obstetrics and Gynaecology*, **83**, 760.

Wu, A., Chanarin, I., and Levi, A. J. (1974): *Lancet*, **1**, 829.