Prevention of Folate Deficiency by Food Fortification

VI. THE ANTIMEGALOBLASTIC EFFECT OF FOLIC ACID-FORTIFIED MAIZE MEAL

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SUMMARY

The antimegaloblastic effect of cooked folic acid-fortified maize meal was tested by feeding the meal to 5 lactating patients with folate-deficient megaloblastic anaemia. The dose of folic acid in the meal before cooking varied from 100 to 500 μg daily. One patient who received 100 μg daily had a suboptimal haematological response. Meal containing 300 μg and 500 μg daily produced optimal haematological response in all 4 patients so treated, and there was no secondary reticulocyte response in the 2 patients subsequently given pharmacological doses of folic acid. It is concluded that the fortified meal after cooking is an effective antimegaloblastic agent.

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There is much interest in food fortification to prevent nutritional deficiencies. As maize, which is poor in folate,[1] is a staple item of the diet of millions of people in developing countries, and as folate deficiency is common in these peoples, we are investigating the use of folic acid-fortified maize meal (FAMM) in the prevention of folate deficiency. Feeding FAMM to pregnant subjects results in a rise in the *Lactobacillus casei* folate activity in their serum and red cells.[2] This indicates that the FAMM provides a form of folate which can be utilised by bacteria, but while this is suggestive, it does not prove that this folate is biologically available to man. For this reason, the efficacy of FAMM as a therapeutic agent in patients with folate-deficient megaloblastic anaemia has been evaluated.

PATIENTS AND METHODS

The antimegaloblastic effect of the cooked meal was tested by means of a conventional therapeutic trial carried out in 5 patients with folate-deficient megaloblastic anaemia. All were adult females who were admitted to hospital with severe anaemia occurring during lactation, and they continued to breast-feed throughout their stay in hospital. The mean period between parturition and admission to hospital was 114 days (range 49 - 189 days). The laboratory findings on admission are summarised in Table I.

On admission to hospital, the patients received the usual hospital diet for periods of 3 - 8 days until the diagnosis of folate deficiency was established by appropriate laboratory investigations. The diet was free of uncooked green leafy vegetables and contained copious quantities of maize meal. This diet did not induce haematological response in patients with folate-deficient megaloblastic anaemia, who were followed for periods of up to 30 days.[3-5]

Patients 2 and 3, who were the most severely anaemic, were given transfusions of whole blood at the time of admission to hospital.

After the diagnosis of folate deficiency had been established, the maize meal in the hospital diet was supplemented with the FAMM. The daily dose of folic acid added to the maize meal before cooking was 100 μg in the case of patient 1, 300 μg in the case of patients 2 and 3, and 500 μg in the case of patients 4 and 5. The maize meal was fortified with pteroylglutamic acid in proportions of one daily dose for each 30 g of maize meal (dry weight). The mixture was cooked in boiling water for 30 - 45 minutes to form a stiff paste, and was eaten with the usual hospital meals each day. We have found that folic acid resists destruction by boiling in water for two hours.[6]

In patients 4 and 5, an attempt was made to elicit a secondary reticulocyte response when the reticulocyte counts had returned to stable levels after feeding the FAMM. These patients were given 15 mg folic acid and 240 mg elemental iron (as ferrous sulphate) daily by mouth.

Standard laboratory methods were used. Haemoglobin, cell counts and red cell indices were measured with a Coulter S counter. Serum folate[7,8] and red cell folate[9] were assayed with *L. casei*, and serum vitamin B_{12} measured by a radio-isotope dilution method.[10]

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TABLE I. RESULTS OF HAEMATOLOGICAL INVESTIGATIONS IN 5 PATIENTS ON ADMISSION TO HOSPITAL (IN ALL PATIENTS THE BLOOD SMEAR SHOWED OVAL MACROCYTES AND HYPERSEGMENTED NEUTROPHILS, AND THE BONE MARROW WAS MEGALOBLASTIC)

	Patient No.				
	1	2	3	4	5
Haemoglobin (g/100 ml)	4,7	3,4	3,7	5,1	4,4
Red cell count (million/μL)	1,30	0,79	1,00	1,25	1,25
Haematocrit (%)	15,3	10,1	11,8	15,3	13,7
Mean cell volume (μm^3)	118	128	118	122	107
Mean cell haemoglobin (pg)	36,2	43,0	37,0	40,8	34,4
Mean cell haemoglobin concentration (%)	30,7	33,7	31,4	33,3	32,1
Leucocyte count/μL	4 800	5 000	2 700	9 800	7 400
Platelet count/μL	30 000	40 000	54 000	85 000	188 000
Reticulocyte count (%)	3,0	2,8	0,6	1,0	3,4
Serum folate (ng/ml)	0,7	1,0	1,2	0,6	0,6
Red cell folate (ng/ml)	48	100	116	12	84
Serum iron (μg/100 ml)	86	134	216	195	241
Serum unsaturated iron-binding capacity (μg/100 ml)	127	175	57	77	31
Serum vitamin B_{12} (pg/ml) (normal range 400 - 1 020 pg/ml)	402	505	554	1 205	1 068
Serum lactic dehydrogenase (units) (normal range 90 - 200)	6 130	4 565	3 110	2 490	2 630

RESULTS

The daily red cell and reticulocyte counts of the 5 patients are depicted graphically in Figs 1 - 5. None of the patients had a reticulocyte response while receiving the hospital diet only, but all showed haematological response after feeding with FAMM. In order for a haematological response in a patient with megaloblastic anaemia to be rated as optimal, three criteria should be satisfied, viz. the reticulocyte peak should occur between days 5 and 8,[11] reach certain levels,[32] and the red cell count must rise at a specified rate.[33] The height of the reticulocyte peak and the rate of rise in red cell count are related to the severity of anaemia. According to these criteria, all the patients in the present study showed an optimal response, except for patient 1 who received the lowest dose of folic acid in FAMM (100 μg/day). In this patient the height of the reticulocyte peak and the rate of rise in the red cell count were suboptimal.

Neither of the 2 patients who received pharmacological doses of folic acid and iron showed a secondary reticulocyte response, after the initial response to the FAMM.

Four patients were thrombocytopenic at the start of the trial, and all had normal platelet counts by day 8. The single patient with a leucopenia on day 0 had a normal white cell count on day 3 of the trial.

DISCUSSION

None of the patients in the present study showed a rise in reticulocyte count during the period in hospital before

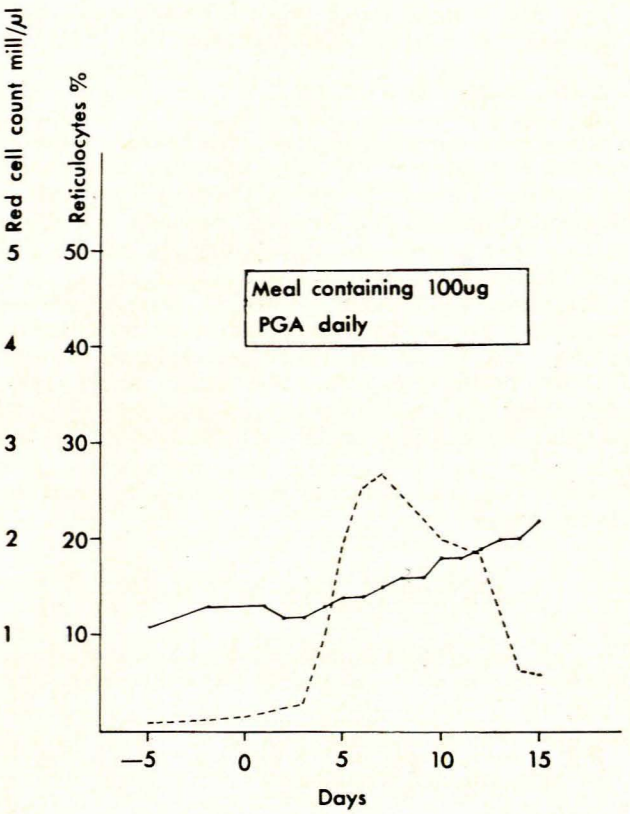

Fig. 1. Reticulocyte response (broken line) **and rise in red cell count** (solid line) **in a patient with folate-deficient megaloblastic anaemia after eating maize meal fortified with 100 μg folic acid/day.**

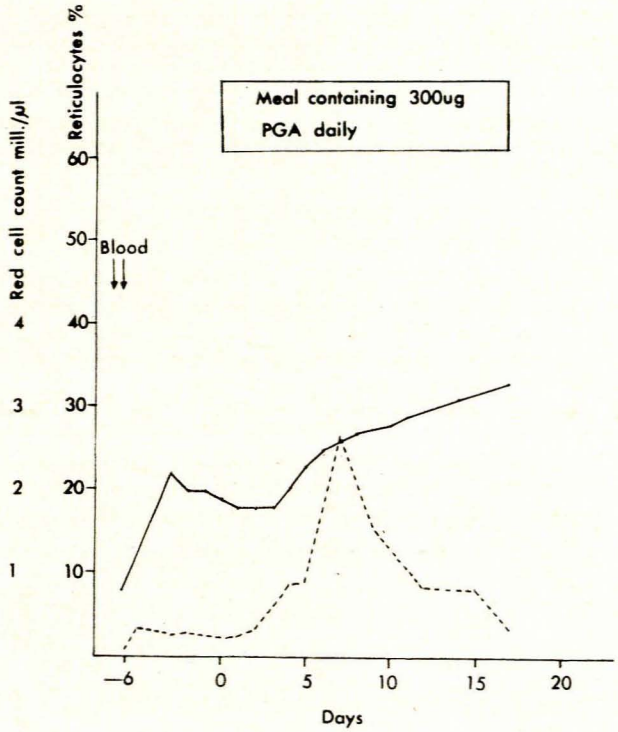

Fig. 2. Reticulocyte response (broken line) **and rise in red cell count** (solid line) **in a patient with folate-deficient megaloblastic anaemia after eating maize meal fortified with 300 µg folic acid/day.**

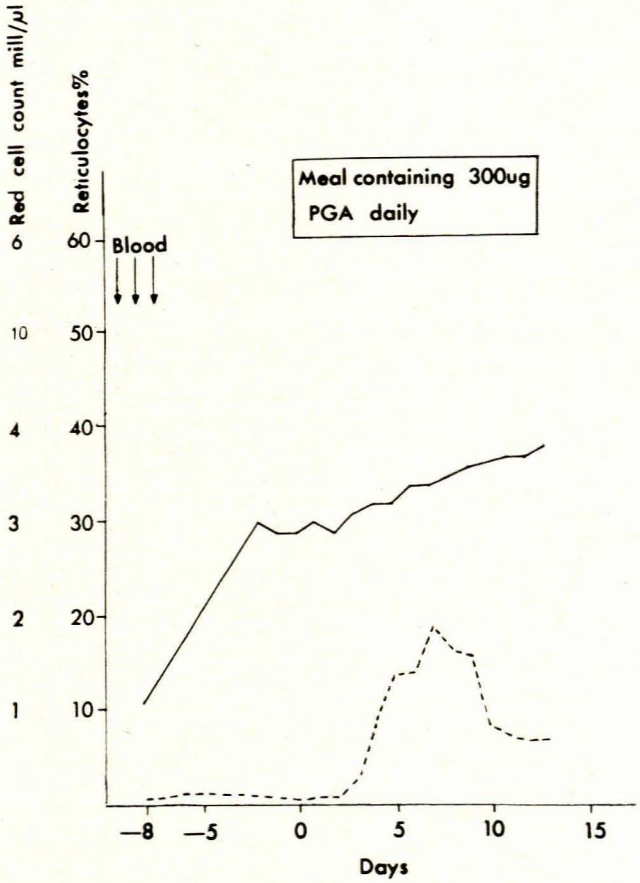

Fig. 3. Reticulocyte response (broken line) **and rise in red cell count** (solid line) **in a patient with folate-deficient megaloblastic anaemia after eating maize meal fortified with 300 µg folic acid/day.**

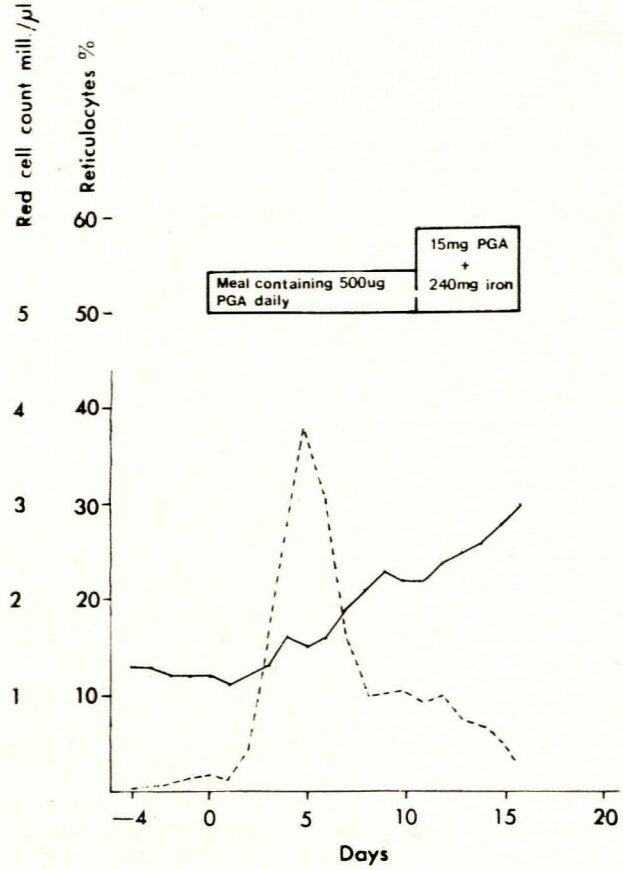

Fig. 4. Reticulocyte response (broken line) **and rise in red cell count** (solid line) **in a patient with folate-deficient megaloblastic anaemia after eating maize meal fortified with 500 µg folic acid per day. There was no secondary reticulocyte response to pharmacological doses (15 mg/day) of folic acid.**

they received FAMM. This is in accord with previous experience with the hospital diet,[8-5] which contains insufficient amounts of folate to induce a haematological response.

Patient 1, in whom the haematological response was suboptimal, received FAMM containing 100 µg folic acid daily before it was cooked. Failure to induce optimal haematological response reflects the fact that this dose is inadequate to meet the folate requirement of the lactating patient, which has been estimated to be approximately 300 µg/day.[14]

The failure to induce a secondary reticulocyte response with pharmacological doses of folic acid in the 2 patients so studied (Nos. 4 and 5) is further evidence of the optimal nature of the response induced by the FAMM containing 500 µg folic acid daily.

In patients 2 and 3, the therapeutic trial followed the administration of blood transfusions. It has been suggested that transfusions may induce a 'spontaneous' response in a patient with megaloblastic anaemia.[15] However, in a previous study, 3 lactating patients with folate-deficient megaloblastic anaemia who were transfused did not have reticulocyte responses when followed for periods of up to 17 days.[4]

The results of these trials indicate that cooking folic acid in the presence of maize meal does not alter its biological efficacy as an antimegaloblastic factor in man.

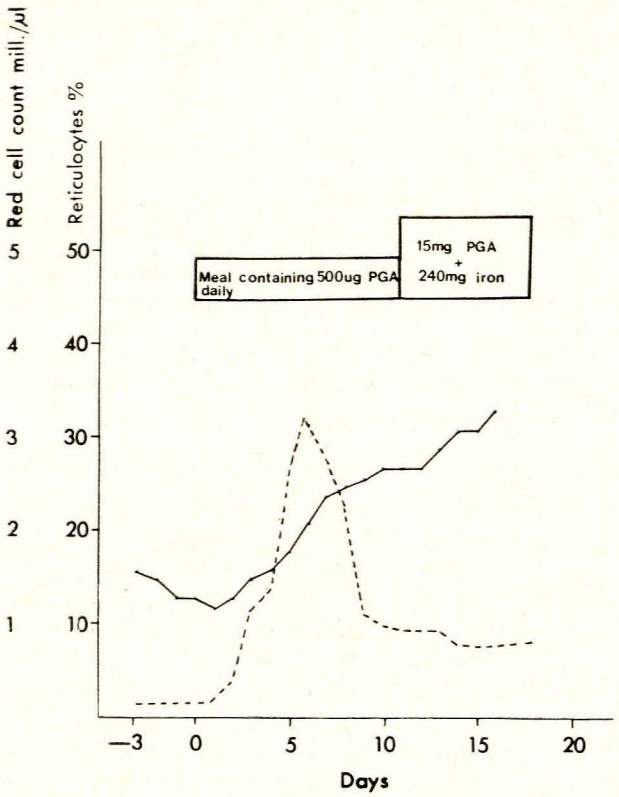

Fig. 5. Reticulocyte response (broken line) **and rise in red cell count** (solid line) **in a patient with folate-deficient megaloblastic anaemia after eating maize meal fortified with 500 µg folic acid per day. There was no secondary reticulocyte response to pharmacological doses (15 mg/day) of folic acid.**

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