Biochemical aspects of malabsorption in marasmus

BY H. C. MEHTA[1], A. S. SAINI[1], HARJIT SINGH[2] AND P. S. DHATT[2]

[1]*Department of Biochemistry and* [2]*Department of Paediatrics, Medical College and Hospital, Rohtak, Haryana 124001, India*

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1. Sixty marasmic children and fifteen normal age-matched controls were investigated for the absorption of fats and proteins. Their duodenal juice samples were also analysed for bile salts and microflora.

2. A significant amount of malabsorption, with respect to both the dietary ingredients, was observed in the majority of the marasmic children.

3. The levels of conjugated bile acids in the duodenal juice samples of marasmic children were significantly lower as compared with those of normal controls, while the reverse was true for free bile acids and bacterial counts.

4. Significant correlations were observed between bile acid levels and fat absorption and also between microflora and free bile acids.

5. The pathophysiology of malabsorption in marasmus is discussed in the light of these findings.

Protein–energy malnutrition (PEM) is, even today, a major public health problem over large areas of the world. Apart from inadequate dietary intake, digestive and absorptive defects may constitute important factors in the aetiology of PEM. It is, however, difficult to say whether these defects are the cause or the effect of malnutrition.

Many alterations in the gastrointestinal tract of malnourished children have been reported. These include gastric mucosal atrophy and achlorohydria (Gracey *et al.* 1977), abundance of bacteria and yeast in gastric and duodenal contents (Mata *et al.* 1972), pancreatic atrophy (Veghelyi *et al.* 1950; Thompson & Trowell, 1952; Barbezat & Hansen, 1968), gastrointestinal mucosal atrophy (Brunser *et al.* 1968), deficiency of intestinal disaccharidases (Gurson & Saner, 1969; Berkel *et al.* 1970; James, 1971) and decrease in conjugated bile acids (Schneider & Viteri, 1974). These derangements may result in impaired digestion and absorption so frequently observed in malnourished children. However, the status of gastrointestinal functions in marasmus is still controversial.

The present work aims at studying the absorption of major dietary ingredients together with duodenal juice analysis in order to understand the pathophysiology of malabsorption in marasmic forms of PEM.

MATERIALS AND METHODS

Sixty marasmic children admitted to the Paediatrics department of Medical College Hospital, Rohtak, constituted the case material for the present study. The weights of these children were less than 60% of 50th percentile of Boston standards (Dugdal, 1971); they had no oedema, were free from any major infection and were in the age group of 9–42 months. Diarrhoea, if present, was brought under control before starting the investigations. Fifteen normal children, weighing more than 80% of 50th percentile of Boston standards and of the same age group, formed the control group. The following investigations were done in all the children, except 24 h faecal fat excretion which was done in twenty of the sixty marasmic children.

Absorption of proteins was studied by giving 1·5 g casein/kg body-weight in the form of calcium caseinate and estimating proline in blood before and 2 h after casein administration (Gould & Schwachman, 1956).

Faecal fat excretion (24 h) was studied in twenty marasmic and fifteen normal children using the method of Vande Kamer *et al.* (1949). For 5 d before the fat excretion studies the children were kept on a diet containing about 50 g fat/d. Lipid absorption was also studied in all the children, using the method of Jones & diSant Agnese (1963).

Duodenal contents were aspirated in clean sterile tubes after a milk stimulus (Lundh, 1962). The presence of the tube in the duodenum was confirmed by fluoroscopy. A biluminal Miller-Abbot tube was used wherever feasible. In other cases, where it was not possible to pass this tube because of thickness, Ryle's tube was used for duodenal intubation.

Duodenal juice pH was immediately noted using narrow range pH papers (British Drug House, Poole, Dorset) and the juice was stored in the refrigerator at 4° for bile acid analysis and the study of microbes, which were done within 24 h of sample collection.

Duodenal juice was analysed for free and conjugated bile acids by thin-layer chromatography (Anthony & Behar, 1964). Quantitative determination of bile acids was done by comparing the unknown spots with the serial standard spots of bile acids which were chromatographed with duodenal juice samples.

Aerobic organisms in the duodenal juice were studied by the standard loop technique using nutrient agar plates (O'Sullivan *et al.* 1960).

RESULTS AND OBSERVATIONS

Bile acids (*Table 1*)

The mean concentration of conjugated bile acids as well as total bile acids was significantly lower in the duodenal juice samples of marasmic children as compared with the normal children ($P < 0{\cdot}001$). The concentration of free bile acids, however, was more than normal in the duodenal juice samples of marasmic children ($P < 0{\cdot}001$).

Table 1. *Bile acid levels in the duodenal juice samples of normal and marasmic children*
(Mean values and standard deviations)

	Conjugated bile acids (mg/ml)			Free bile acids (mg/ml)		
Children	Range	Mean	SD	Range	Mean	SD
Normal (*n* 15)	2·3–3·8	2·92	0·57	0·12–0·40	0·30	0·06
Marasmic (*n* 54)	0·52–3·4	1·36***	0·68	0·08–1·56	0·60***	0·30

*** $P < 0{\cdot}001$.

Microflora (*Table 2*)

There was a significant increase in the microbial content of duodenal juice samples collected from marasmic children. While only two out of fifteen normal children showed slight growth of aerobes in duodenal juice, thirty out of fifty-four marasmic children showed significant growth in their duodenal juice samples.

Absorption studies (*Table 3*)

Faecal fat excretion (24 h) was significantly higher in marasmic as compared with the normal children ($P < 0{\cdot}01$). Daily fat excretion exceeded 5 g in 60% of the marasmic children. The lipid absorption test also indicated a significant amount of fat malabsorption in marasmic children.

There was a significant negative correlation ($r -0{\cdot}8$, $P < 0{\cdot}001$; $y = 3{\cdot}66 - 0{\cdot}37\ x$) between 24 h faecal fat excretion and conjugated bile acids (Fig. 1) and a significant positive

Table 2. *Aerobic organisms in duodenal juice samples of normal and marasmic children*

Children	Aerobes			Log bacterial count		
	Sterile	Count $< 10^4$	Count/10^4	Range	Mean	SD
Normal (*n* 15)	13	2	—	0–2·0	0·23	—
Marasmic (*n* 54)	24	—	30**†	0–5·5	2·64	2·4

** $P < 0{\cdot}01$.

† Type of organism (number of instances in parentheses): *Escherichia coli* (22), *Pseudomonas* spp. + *E. coli* (3), *Candida albicans* (1), *C. non-albicans* (2), others (2).

Table 3. *Absorption of fats and proteins in normal and marasmic children*

(Mean values and standard deviations)

Children	Protein absorption			Fat absorption				
	Rise in serum proline (μmol/l) after a casein load			24 h faecal fat excretion (g) (*n*20)			Lipid absorption (urine dilution positive for iodine)†	
	Range	Mean	SD	Range	Mean	SD	< 1:4	> 1:4
Normal (*n* 15)	208·5–521·2	330·1	95·5	1·15–2·25	1·65	0·36	1	14
Marasmic (*n* 60)	43·4–460·4	191·1***	86·9	1·50–9·40	4·68**	2·34	33	25

** $P < 0{\cdot}01$, *** $P < 0{\cdot}001$.

† Values expressed as number of cases.

correlation between lipid absorption and conjugated bile acids (r 0·5, $P < 0{\cdot}001$). There was a significant positive correlation between the level of aerobes and free bile acids in the duodenal juice (r 0·39, $P < 0{\cdot}01$) and a significant negative correlation between the level of aerobes and the ratio, conjugated:free bile acids (r −0·48, $P < 0{\cdot}001$).

The rise in serum proline after a casein load was also significantly lower in marasmic as compared with the normal children ($P < 0{\cdot}01$) (Fig. 2). A rise in serum proline above 217 μmol/l was taken as an index of good absorption, between 130 and 217 μmol/l as moderate absorption and less than 130 μmol/l as poor absorption. Accordingly, 37% of the marasmic children revealed good protein absorption as compared with the corresponding value of 93% in normal children. Only 7% of the normal children (one out of fifteen) showed moderate absorption compared with 42% of the marasmic children. The remaining 21% of the marasmic children showed poor protein absorption.

DISCUSSION

Duodenal juice samples of marasmic children contained significantly lower amounts of conjugated bile acids and significantly higher amounts of free bile acids as compared with the normal children. Similar results in malnourished children have been reported by other workers (Mahanabalis *et al.* 1972; Redmond *et al.* 1972; Viteri & Schneider, 1974) and these may be attributed, apart from diminished hepatic synthesis, to excessive bacterial counts observed in marasmic children of the present and other studies (Mata *et al.* 1972; Gracey *et al.* 1977). Deconjugation of bile acids by micro-organisms is a well-known phenomenon (Dawson & Isselbacher, 1960; Donaldson, 1964; Kim *et al.* 1966) and we observed a significant correlation between free bile acids and bacterial counts.

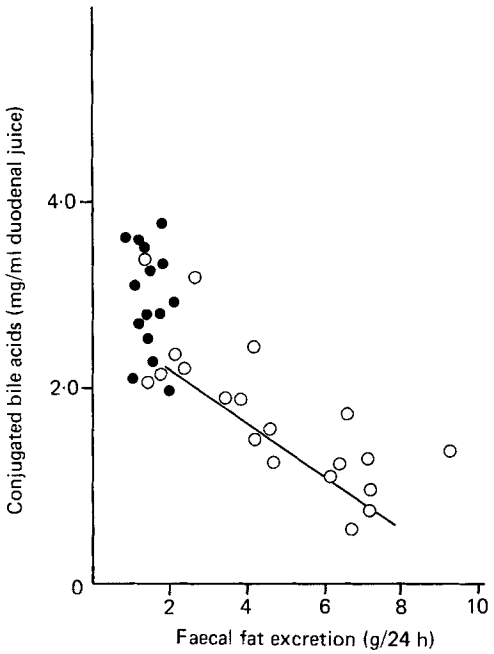

Fig. 1. Correlation between 24 h faecal fat excretion and conjugated bile acids in duodenal juice. $y = 3{\cdot}66 - 0{\cdot}37x$. (●) Normal children, (○) marasmic children.

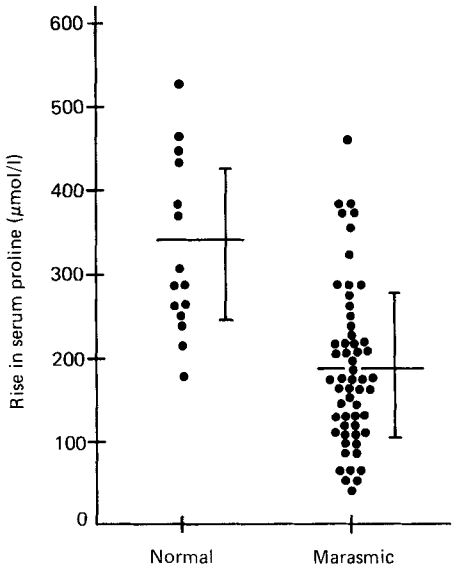

Fig. 2. Rise in serum proline (μmol/l) after a casein load (1·5 g casein/kg body-weight) in normal and marasmic children. Mean values shown by horizontal bars, vertical bars represent standard errors.

There was a significant impairment in the absorption of fats as well as proteins (Table 3) and the results support other reports indicating varying amounts of fat malabsorption in marasmic children (Amin *et al.* 1969; Teotia *et al.* 1969; Gupta *et al.* 1970; Singh *et al.* 1977). Protein absorption, however, has been reported to be normal by most workers (Holeman & Lambrechts, 1955; Hansen, 1956; Robinson *et al.* 1957), although Viteri *et al.* (1973) reported an increased nitrogen excretion in malnourished children and Singh *et al.* (1977) observed a significant amount of protein malabsorption with the technique used in the present study.

Although a number of factors play a role in fat absorption, the most significant of these appears to be the concentration of conjugated bile acids. A decreased concentration of conjugated bile acids results in impaired micellar formation, since the critical micellar concentration of conjugated bile acids is much lower than that of free bile acids (Hoffman & Borgstrom, 1962). It is pertinent to note that there was a significant negative correlation between faecal fat excretion and conjugated bile acids. We also observed low lipase activity and low pH of duodenal juice samples of marasmic children (H. C. Mehta, A. S. Saini, H. Singh and P. S. Dhatt, unpublished results) which may also affect digestion and absorption of fats but these do not appear to be very critical factors in fat absorption. Abnormal mucosal patterns may also contribute to generalized malabsorption. We studied jejunal mucosal patterns in a few cases and noted only mild to moderate alterations.

The original concept that all protein is absorbed in the form of amino acids has radically changed and now it is clear that much protein is absorbed in the form of oligopeptides which are further hydrolysed by brush-border proteases and cytosol dipeptidases (Matthews, 1975). N balance studies take into account mainly the intraluminal events which are hydrolysis of proteins to oligopeptides and subsequent uptake of oligopeptides by the mucosal cells. Such studies may, therefore, be normal if the previously mentioned intraluminal events are unaffected but amino acids will rise in the blood at a normal rate only if the intramucosal hydrolysis of oligopeptides is also normal. Thus, normal faecal N excretion of various studies can be explained by presuming that intraluminal hydrolysis of proteins is unimpaired but intramucosal hydrolysis of oligopeptides and release of amino acids is diminished. Deficiency of intestinal disaccharidases, in fact, is known to occur in marasmic children (Kumar *et al.* 1971).

The present work suggests that malabsorption is a common phenomenon in the marasmic type of PEM. Though it may be the result of an interplay of a number of factors such as diminished pancreatic functions, intestinal microflora and mucosal changes, the concentration of conjugated bile acids seems to be critical in fat absorption and that of mucosal oligopeptidases and dipeptidases in protein absorption.

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