

# Plasma somatomedin activity in protein calorie malnutrition

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**SUMMARY** Somatomedin activity was assayed in the plasma of children suffering from protein calorie malnutrition by a bioassay using rat cartilage and expressed as sulphate uptake ratio. The sulphate uptake ratio was particularly reduced in kwashiorkor. In marasmus there was a slight reduction and the levels were still in the normal range. Plasma growth hormone (GH) levels were raised in kwashiorkor but were in the normal range in marasmus. Reduction in sulphate uptake ratio was observed only when plasma albumin levels were less than 2.5 g/100 ml (25 g/l). A rise in plasma GH was also observed but only below this threshold level.

Some of the actions of growth hormone (GH) on cartilage are believed to be mediated through GH-dependent serum factors—the somatomedins (Daughaday, 1971). The liver is one of the known sites of somatomedin generation that has been shown to be stimulated by GH (McConaghey, 1972).

Raised GH levels have been observed in kwashiorkor (Raghuramulu and Jaya Rao, 1974), a condition in which the concentrations of various proteins synthesised in the liver, including albumin, are markedly decreased. A lowered level of serum somatomedin may act as a feedback to stimulate excess production of GH in this condition. Plasma somatomedin is generally measured by bioassays, in which the uptake of either  $\text{Na}_2^{35}\text{SO}_4$  or  $^3\text{H}$ -thymidine by cartilage in the presence of serum is determined. To test whether any correlation exists between plasma GH levels and somatomedin activity in protein calorie malnutrition, the ability of serum from children suffering from such malnutrition to stimulate  $\text{Na}_2^{35}\text{SO}_4$  uptake by rat cartilage was studied.

## Patients and methods

Eight children suffering from kwashiorkor and 7 marasmic children were studied. They conformed to descriptions given earlier (Raghuramulu and Jaya Rao, 1974). Investigations were carried out immediately on admission to hospital and after 3 or 4 weeks

of nutritional rehabilitation on diets which provided 200 kcal (0.836 MJ) and 4 g protein/kg daily. A sample of venous blood was obtained after an overnight fast, under resting conditions. Blood was collected under heparin, plasma separated, and stored at  $-20^\circ\text{C}$ .

The bioassay was carried out as described by Yde (1968) using 21–27 day-old male albino rats, fasted for 48 hours before the assay. The cartilage pieces were however not subjected to acid hydrolysis but digested with papain (Alford *et al.*, 1972). The radioactivity in the hydrolysate was measured in a Packard liquid scintillation counter, using Bray's scintillating solution.

The assay was carried out by a symmetrical 4-point bioassay using both reference (pooled plasma from healthy adults) and test plasma at 10% (0.07 ml) and 20% (0.14 ml) of the total incubation volume (0.7 ml). Each concentration of the plasma was assayed in triplicate. Radioactivity was expressed per mg dry weight of the cartilage. The ratio of the radioactivity obtained with the test plasma to that obtained with the reference plasma was calculated and the average of the 6 observations was taken as the sulphate uptake ratio. Plasma samples of each child obtained on admission and after nutritional rehabilitation were assayed simultaneously, using the same rat to minimise interanimal variations.

In 7 independent assays normal plasma was used at 0, 2.5, 5, 7.5, 10, 12.5, 15, 17.5, 20% v/v concentrations and log dose response curves were constructed. The relationship was found to be linear up to a concentration of 20% v/v and the index of precision ( $\lambda$ ) was  $0.18 \pm 0.020$  (mean  $\pm$  SE).

Plasma GH was estimated by the double antibody

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technique of Pennisi (1968), the materials used being similar to those described earlier (Raghuramulu and Jaya Rao, 1974). Plasma albumin was estimated by the biuret method (Wolfson *et al.*, 1948).

**Results**

The results of the study are presented in the Table. The mean sulphate uptake ratio in children suffering from kwashiorkor was significantly lower than that observed in normal children ( $P < 0.001$ ) and the values rose significantly after nutritional rehabilitation ( $P < 0.01$ ). The mean value in marasmic children was not different from that observed in normal children. However, there was a slight but significant increase after treatment ( $P < 0.05$ ).

The mean basal plasma GH level in children suffering from kwashiorkor was significantly higher than that observed in normal children ( $P < 0.01$ ). The levels came down significantly after nutritional rehabilitation ( $P < 0.05$ ). The mean level in marasmic children was not different from that observed in normal children and there was no change after treatment.

Correlations were tested between sulphate uptake ratio and plasma GH, between sulphate uptake ratio and plasma albumin, and between plasma GH and plasma albumin; both groups of children were tested separately before and after treatment. No positive correlations were obtained in any of the groups when tested individually. As the coefficients of correlation for both groups, before and after treatment, were not significantly different the entire data were combined for analysis. A significant correlation was obtained only between sulphate uptake ratio and plasma albumin concentration ( $r = 0.576$ ,  $P < 0.001$ , Figure).

Table Plasma sulphate uptake ratio and GH levels in protein calorie malnutrition

	Sulphate uptake ratio	GH (ng/ml)
Normal children (n = 11)	1.06 ± 0.072	8.4 ± 1.83
Kwashiorkor (n = 8)		
Before treatment	0.61 ± 0.059‡	30.8 ± 8.29†
After treatment	0.97 ± 0.085**	6.8 ± 2.18*
Marasmus (n = 7)		
Before treatment	0.97 ± 0.057	5.2 ± 0.82
After treatment	1.09 ± 0.056*	3.9 ± 1.06

Before treatment v. after treatment

\*  $P < 0.05$ , \*\*  $P < 0.01$ .

Normal v. children with protein calorie malnutrition

†  $P < 0.01$ , ‡  $P < 0.001$ .

**Discussion**

Data show that the plasma sulphate uptake ratio is lowered in kwashiorkor. As bioassay of somatomedin involves the stimulation of tissue sulphate uptake in the presence of plasma, the present study may be considered to indicate that plasma somatomedin activity may be lowered in kwashiorkor, an observation in line with that reported by Grant *et al.* (1973). Our data also show additionally that the sulphate uptake ratio is not altered in marasmus. The slight increase after treatment indicates that the reduction, if any, is only marginal. Van den Brande and Du Caju (1973) studied somatomedin activity in malnourished children and although they combined the data obtained from marasmic children with those from children with kwashiorkor, it would appear that both groups had low levels of activity. Their data also indicate the probable presence of a somatomedin inhibitor in the plasma of marasmic children. This was not tested in the present series. The normal sulphate uptake observed in marasmic children could probably be taken as evidence against the presence of such an inhibitor. The presence of an inhibitor in kwashiorkor however is not ruled out.

The finding that plasma GH levels are raised in kwashiorkor but normal in marasmus agrees with our earlier findings (Raghuramulu and Jaya Rao, 1974). There is a consensus regarding the high GH levels in kwashiorkor (Pimstone *et al.*, 1966; Beas *et al.*, 1971; Godard, 1973; Parra *et al.*, 1973; Suskind *et al.*, 1973) but observations on marasmus are discordant (Pimstone *et al.*, 1968; Beas *et al.*, 1971; Godard, 1973; Parra *et al.*, 1973; Suskind *et al.*, 1973). Circulating levels of several proteins known to be synthesised in the liver are markedly lowered in kwashiorkor (Gopalan, 1968). Liver is also one of the sites of somatomedin generation (McConaghey, 1972; Williams and Hughes, 1974). The lowering of somatomedin

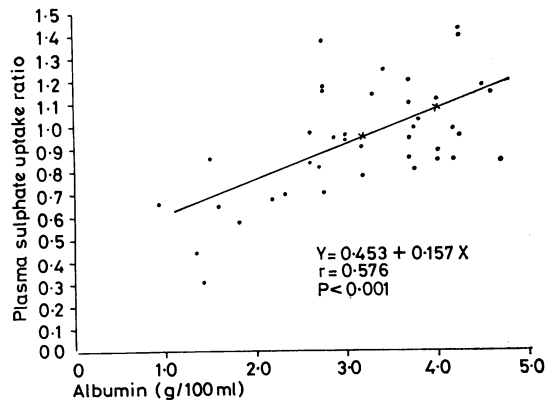


Figure Relationship between plasma sulphate uptake ratio and plasma albumin level.

activity in kwashiorkor might therefore be due to its impaired synthesis, as was suggested by Grant *et al.* (1973).

It has been suggested that plasma amino-acid levels may determine plasma GH concentration in protein calorie malnutrition (Suskind *et al.*, 1973; Jaya Rao, 1974). The possibility that a decrease in somatomedin feedback, either on the pituitary or on the hypothalamus, could also stimulate excess GH production needs to be tested. The lack of correlation between the two substances at the outset precludes such a possibility. On the other hand, it was observed that in all children (except one) who had plasma albumin levels below 2.5 g/100 ml (25 g/l) the sulphate uptake ratio was below 0.7 (Figure). It has earlier been observed that plasma GH levels were raised only when the plasma albumin level fell below this concentration (Raghuramulu and Jaya Rao, 1974). The data of other workers (Pimstone *et al.*, 1968; Samuel and Deshpande, 1972; Lunn *et al.*, 1973) also support the latter observation. The results of the present study indicate that the reduction in sulphate uptake ratio as well as raised plasma GH levels are observed only below a certain threshold level of plasma albumin. They however, do not indicate whether somatomedin activity could be the primary determinant of the raised plasma GH levels in kwashiorkor.

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