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Blood Plasma Levels of Cortisol, Insulin, Growth Hormone and Somatomedin in Children with Marasmus, Kwashiorkor, and Intermediate Forms of Protein-Energy Malnutrition (41222)

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Abstract. Plasma insulin, growth hormone, somatomedin, free and bound cortisol, and serum albumin were measured in human infants with protein-energy malnutrition and in age-matched control subjects. Insulin and somatomedin were reduced, growth hormone and free and total cortisol were elevated in malnourished infants. Free cortisol and somatomedin were negatively correlated ($r = -0.78$) and somatomedin and percentage weight deficit were negatively correlated in marasmus ($r = -0.62$) and kwashiorkor ($r = -0.68$). It is suggested that elevated plasma-free cortisol as well as depressed plasma insulin in protein-energy malnourished infants is partially responsible for the depressed plasma somatomedin and reduced protein synthesis and growth in children.

Nutritional status is an important determinant of serum somatomedin (1–6) and other hormone (7) concentrations. The growth-retarding effects of abnormally high levels of circulating glucocorticoids have been reported by several workers (8–12). Although there seems to be agreement that physiologically high levels of circulating glucocorticoids cause growth retardation, there are divergent views as to the mode of action. Some workers (9, 10) reported that excess glucocorticoids affect the morphology and metabolism of chondrocytes of the tibial epiphyseal plate which are intimately related to the continuing growth process, while Elders *et al.*, (11, 12) postulated that glucocorticoids cause growth retardation by inhibiting somatomedin synthesis. Phillips *et al.* (8), however, reported that glucocorticoid-induced growth retardation may result from any of these four mechanisms—a decrease in growth hormone release, a decrease in growth hormone-induced somatomedin generation, a decrease in somatomedin action on cartilage, or a direct inhibitory effect on cartilage. Somatomedins are intimately involved with the growth process (13–15); this study was designed to evaluate the relationships among serum albumin and plasma growth hormone, insulin, somatomedins, and free and total cortisol in children with clinical evidence of protein-energy malnutrition (PEM).

Experimental Procedure. Sixteen children between 8 and 34 months of age and suffering from varying degrees of PEM were available for the study. Subjects were randomly selected during a 1 week period from a group of children attending the daily outpatient pediatric clinics of the General and Teaching Hospitals in Enugu, Nigeria. All of the children included in this study were medically diagnosed as malnourished by the consulting pediatricians. Control subjects aged between 18 and 24 months were selected from among healthy children of the hospital staff. The age, weight, and height of each subject was recorded. The Wellcome classification of protein-energy

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malnutrition (16) was used and the subjects were classified as having marasmus, kwashiorkor, or intermedite forms of PEM.

Blood samples were collected from each subject, after clinical examination, into heparinized and plain tubes. Samples were centrifuged within 1 hr to obtain plasma and serum, respectively, and stored at -30° until analyzed for insulin, growth hormone, somatomedin, and free and bound cortisol. Blood samples were collected from the experimental subjects after unknown periods of fast. At both the General and Teaching Hospitals, patients had to wait for several hours before being attended by a physician so that blood samples collected from these subjects were essentially fasting.

Serum albumin was measured by the Biuret method of Gornall *et al.* (17). Plasma insulin was measured by the double-antibody radioimmunoassay method of Hales and Randle (18) with all reagents supplied in a kit (Amersham, Arlington Heights, Ill.). Growth hormone was measured by the double-antibody radioimmunoassay described by Melani *et al.* (19) with all reagents supplied in a kit (Serone Labs. Inc., Braintree, Mass.). Serum somatomedin activity was measured by non-dose–response techniques because of the low volumes available. Sulfate incorporation by hypophysectomized rat control cartilage exposed to 5% serum (v/v) was determined in 2-day incubations (20). The bioassay was used instead of a radioimmunoassay to measure inhibitors as well as somatomedins. Somatomedin activity is expressed as micrograms of sulfate uptake per 100 mg dry cartilage in 5% serum. Total serum cortisol was measured by the use of the dextran-coated charcoal radioimmunoassay as described by Krey *et al.* (21). Because of the apparently high levels of circulating cortisol in humans, serum samples were diluted 1:20 and with 25 μl of the diluted serum used in the assay; the actual volume of serum assayed was 1.25 μl. Free cortisol concentrations were determined by a modification of the assay for transcortin-bound and -unbound cortisol developed by Ballard and Carter (22). This modification involved the use of gel filtration instead of charcoal used in the original method to separate bound cortisol from free. Data were subjected to analysis of variance, and differences were identified by a range test (23).

Results and Discussion. Table I gives a summary of anthropometric measurements and serum albumin levels of both control and experimental subjects. Except for patients AD3 and AD5, serum albumin levels in children with intermediate forms of PEM were within the range of 3–4.5 g/dl which is considered normal at these hospitals (D. O. Tabansi, personal communication). Serum albumin levels in children with PEM were significantly lower ($P<0.001$) than those of controls. However, the mean level in the marasmic children was higher but not significantly different from that observed in children with kwashiorkor. These results agree with reports from other human and animal studies (24–28). Although one of the clinical signs of kwashiorkor is reduced serum albumin, the low end of the normal range appears to vary from one study to another. Whitehead *et al.* (24) reported that 3.0 g/dl albumin is the critical concentration below which abnormalities appear in blood hormone levels and levels of other blood constituents. In contrast, Leonard (26) observed a critical level of 2.0 g/dl. The results of this study suggest that the critical level of serum albumin depends on what biochemical changes are being studied (Table II). Insulin and somatomedin levels were very low in some patients with serum albumin levels of 3.0 g/dl and above. However, growth hormone levels corresponded to changes in serum albumin concentration and were higher in malnourished children with serum albumin concentration of 3.0 g/dl and less.

The response of serum somatomedin activity to PEM was as dramatic as the change in insulin concentrations. Mean somatomedin activity in the children with PEM was significantly less than control values ($P<0.005$); however, the mean in children with kwashiorkor was not significantly different from that of the marasmic children. It is recognized that the bioassay procedure for somatomedin measures in-

TABLE I. ANTHROPOMETRIC MEASUREMENTS AND SERUM ALBUMIN LEVELS IN CONTROL AND EXPERIMENTAL SUBJECTS SUFFERING FROM MARASMUS, KWASHIORKOR, AND INTERMEDIATE FORMS OF PROTEIN-ENERGY MALNUTRITION

Subject	Age (months)	Weight (kg)	Weight for age[a] (%)	Height (cm)	Height for Age[a] (%)	Serum albumin (g/dl)
Controls						
C1	24	11.7	90.5	87.5	100.0	5.1
C2	24	11.5	90.0	85.2	97.0	4.7
C3	18	11.5	100.0	83.0	100.0	5.1
C4	20	11.8	100.0	85.0	100.0	4.8
	$\bar{X}$ 21.5					
Marasmus						
AD1	19	6.5	60.0	76.0	92.0	3.0
AD8	14	6.1	60.0	73.0	94.0	3.2
P11	24	8.0	65.0	78.0	90.0	3.6
	$\bar{X}$ 19.0					
Kwashiorkor						
AD7	19	7.4	65.0	75.0	87.0	2.4
P7	24	8.0	65.0	78.0	88.0	2.5
P10	24	9.0	70.0	78.5	90.0	2.0
	$\bar{X}$ 22.3					
Intermediate forms						
AD4	21	8.3	70.0	75.0	88.0	3.2
P1	22	8.0	65.0	73.0	85.0	3.9
P6	34	9.0	65.0	79.5	85.0	4.0
P2[b]	15	9.0	85.0	72.5	93.8	3.5
AD2	8	6.0	70.0	66.0	95.0	3.1
P9[b]	18	10.0	90.0	74.5	91.5	3.5
P8	17	8.0	75.0	73.0	90.0	4.1
AD3	18	7.5	65.0	73.0	90.0	2.7
AD5	10	6.0	65.0	70.0	97.0	1.7
P3	16	7.0	65.0	73.0	92.1	3.2

[a] Expected weight and height for age is equivalent to the Boston 50th percentile which is used as the standard.

[b] These subjects had slight edema of the feet and abdominal distention.

hibitors as well as somatomedins. The results show that decreased net somatomedin activity (somatomedins plus inhibitors) contributes to poor growth in malnourished children. Serum somatomedin concentration is greatly influenced by nutritional status (1–6, 22–24); dietary protein and energy deficiency may lead to impaired growth through a decrease in circulating somatomedin or a decrease in cartilage re-

TABLE II. SERUM ALBUMIN AND PLASMA HORMONE CONCENTRATIONS IN CHILDREN WITH MARASMUS, KWASHIORKOR, AND INTERMEDIATE FORMS OF PROTEIN-ENERGY MALNUTRITION

	N	Serum albumin g/dl	Insulin μU/ml	Growth hormone ng/ml	SM[a] (μg SO_4)	Total cortisol (μg/dl)	Free cortisol (%)
Control	4	4.9 ± 0.2	17.4 ± 8.9	0.31 ± 0.4	10.0 ± 4.2	9.1 ± 4.5	18.7 ± 7.0
Marasmus	4	3.1 ± 0.5	1.4 ± 1.0	13.4 ± 6.8	1.0 ± 0.8	28.5 ± 9.6	44.2 ± 15.8
Kwashiorkor	3	2.5 ± 0.6	1.6 ± 0.9	12.6 ± 8.2	2.2 ± 2	16.2 ± 1.4	29.8 ± 20.6
P		0.001	0.05	0.05	0.005	0.01	0.05

[a] Somatomedin (SM) activity is expressed as micrograms of sulfate per 100 mg cartilage.

sponsiveness to somatomedin. Jaya Rao (29) hypothesized that impaired hepatic function consequent to fatty infiltration may be responsible for reduced synthesis and a decrease in circulating somatomedin in PEM. Fatty infiltration of the liver is a feature of kwashiorkor which is not common in marasmus. The data on somatomedin activity in marasmus and kwashiorkor (Table II) and the observation by Hintz *et al.*, (2) that somatomedin levels were the same in marasmus, kwashiorkor, and marasmic kwashiorkor do not support this hypothesis. Phillips *et al.* (4, 8) and Daughaday *et al.* (30) suggested that the defect in somatomedin production in cases of PEM could be partly due to insulin deficiency as well as nutritional depletion. No correlation was found between insulin concentrations and serum somatomedin activity in this study. The growth retardation in these children may have been caused by one or both of these factors: (i) decrease in cartilage responsiveness to somatomedin stimulation or (ii) decrease in circulating somatomedin due to decreased synthesis.

Physiologically high levels of circulating cortisol significantly inhibit the stimulatory action of somatomedin on epiphyseal cartilage sulfate incorporation by causing a derangement in the morphology and metabolism of the chondrocytes (31–34) and also inhibit somatomedin synthesis (4, 5, 34). Mean levels of both total and free cortisol were significantly ($P<0.01$) higher in the three groups of malnourished children compared to the controls (Table II), while the mean level in the marasmic children was significantly higher than that of children with kwashiorkor ($P<0.001$). However, no significant correlation was found between either somatomedin activity and total cortisol or percentage weight deficit and total cortisol in the three groups. In contrast, in both the marasmic children and children with kwashiorkor, a close relationship was found between free cortisol and somatomedin ($r = -0.78, -0.99$) and free cortisol and percentage weight deficit ($r = 0.93, 0.73$). A high correlation was also found between somatomedin and percentage weight deficit in marasmus ($r = -0.62$) and kwashiorkor ($r = -0.68$).

Leonard (26) reported that the mean percentage free cortisol in children being treated for kwashiorkor fell from an initial value of 35.1 to 18.4% after treatment. In another study (25), a mean percentage unbound cortisol of 56% was observed in a group of malnourished children compared with 33% in the control subjects. The data on mean percentage free cortisol from this study confirm these reports and the results implicate both serum somatomedin deficiency and high serum-free cortisol in the growth depression observed in the malnourished children. However, the deficiency condition did not seem to have affected linear growth in the malnourished subjects. The lack of correlation between somatomedin and percentage height deficit, and free cortisol and percentage height deficit suggests that the growth retardation in these children could not have been as a result of a decrease in cartilage responsiveness to somatomedin stimulation which would affect linear growth. The growth retardation in children suffering from PEM is associated with a rise in plasma-free cortisol and growth hormone and a decrease in plasma insulin concentration. Somatomedin potency as measured with a single-dose bioassay, although only semiquantitative, appears to be a valid indicator of biological relevance. Plasma somatomedin activity was only one-third that of controls despite elevated growth hormone concentration. The possible role of insulin as well as of free cortisol must be recognized (35, 36). It is suggested that the high plasma-free cortisol as well as reduced insulin was partially responsible for the depressed stimulatory action of somatomedin on protein synthesis and growth.

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