Zinc Status, Body Composition, and Maturation in Aboriginal Boys

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Anthropologists believe that the aboriginal people migrated to Australia some 40,000 to 50,000 years ago. It is not known from whence they came, but they do have variant forms of the enzyme carbonic anhydrase in their red blood cells (1) a finding discovered only in certain areas of northwest India. After the coming of the white man 200 years ago, the people were eventually placed in missions. Thus, from the hunter–gatherer–nomadic life, which required no special sanitation or growing of crops, they became fixed in small dwellings, with poor sanitation and generous supplies of white flour, salt, and refined sugar. They no longer were exposed to their traditional foods, such as kangaroo meat, goanna, snakes, lizards, witchety grubs, birds, seeds, nuts, and sugar ants and wild honey (their only source of sweetness). Only on occasion now do they “go bush” and obtain these foods. The stipend from the government (fortnightly) is usually expended within a few days. These funds are gambled. Pecuniary rewards have never been part of their culture.

In the missions, at the present time, the children are noted to grow poorly—usually following the fifth percentile of weight for age—and are subject to gastrointestinal infections and, in particular, bacterial, viral, and parasitic infestations from time to time. Their supply of red meat can be limited to a few days every 14 days, depending also on management of the missions and the political climate. Examples of kwashiorkor or marasmus are unusual; the children appear mainly undersized for age, are generally happy and playful, and do not show evidence of hunger.

During the past decade, our investigations of aboriginal children for body composition by indirect methods revealed that protein reserves were reduced and that many had reduced plasma zinc concentration (2). Subsequent work at yearly intervals in missions in northern Australia revealed that in over 1,000 children, hypozincemia, hypoferremia, and hypercupremia were present in more than 50% of the individuals (3–6). At one mission, Kalumburu, there was a 100% incidence of
hypercupremia (>2 SD above the norm) and an 80% incidence of hypozincemia (2 SD below the norm). For the past 3 years, hair analyses have been studied each year in 200 children (longitudinal study) by Smith et al. (7,8), and the occurrence of low levels of Zn in hair is common.

The present report directs attention toward investigations carried out on a group of 40 to 50 boys 6 to 13 years of age at La Grange Settlement over a 3-year period, 1981 to 1983, during the month of August. La Grange is situated 100 km south of Broome (a coastal town involved in years gone by in the pearling industry). Broome is located in the northwest of the state of Western Australia in the heart of the Kimberley district, on the coast (Fig. 1).

Evidence is reviewed concerning changes in zinc status (blood cells), and consideration is given to new information concerning changes in growth, body composition, and the distribution of water, together with information on water turnover. Also, very recent evidence is presented concerning plasma levels of dehydroisoepiandrosterone sulfate (DHEAS) in 192 children from four coastal missions of the

FIG. 1. Map of Australia showing the portion of the Kimberley in northwest Australia where investigation of zinc status of the people (mainly children) has been carried out. This map is concerned mainly with the coastal region.
This precursor of testosterone is taken as a measure of sexual maturation.

**CLINICAL MATERIAL AND METHODS**

Twenty-five Caucasian children, 6 to 13 years of age, volunteered for study as "controls." These were healthy, normally grown children from Adelaide schools. A total of 57 Caucasian children volunteered for DHEAS studies. From 1981 to 1983, boys aged 6 to 13 years were studied after a thorough clinical examination to exclude any who were sick. Such was unusual. Height and weight were determined as described previously (4).

A blood sample (10 ml) was taken, placed in a centrifuge tube containing a small amount of heparin, and centrifuged at 200×g for 10 min. The red blood cells (RBCs) were resuspended in an equivalent volume (i.e., 10 ml) of Earle's balanced salt solution, and this mixture was layered onto a Hypaque Ficoll medium in a ratio of 5:3. The tube and contents were centrifuged at 200×g for 30 min. The separate bands of lymphocytes and granulocytes were thus separated by the one-step method of Ferrante and Thong (9). These bands were removed and washed in Earle's solution and divided into aliquots for (a) counting, (b) identification of cell types with a cytopsin, and (c) acid digestion prior to zinc analysis.

All reagents were free of zinc in terms of detectability by our equipment. In our hands, no clumping of cells was experienced. All Zn analyses were performed with a Perkin-Elmer atomic absorption spectrometer.

For RBC studies, the cells were hemolyzed and the hemoglobin and Zn measured. For urine studies, creatinine was determined by the method of Hare (10). For measurement of total body water, pure heavy water was used; extracellular volume was determined from the distribution of bromide (Br) corrected for the amount of Br entering cells (11).

After administration orally (in fruit juice) of exactly 30 g of deuterium oxide (D$_2$O) containing a total of 1 g of NaBr, a blood sample was taken at 3 hr. Plasma D$_2$O was determined after distillation by infrared spectrophotometry, with temperature control of the measuring cell. Bromide was measured by the Goodwin colorimetric method. An almost identical result for Br was obtained by proton-induced x-ray emission, using only a few microliters of plasma and strontium as an internal standard. These methods have been restated and discussed in detail elsewhere (12).

The corrected bromide space (CBS) allows for 10% of nonextracellular bromide (RBCs, gastric mucosa, and renal epithelium), and the plasma Br is divided by 0.95 (the Gibbs-Donnan factor) and by 0.93 (percent of water in plasma) to yield the concentration of Br in extracellular fluid: [Br]$_e$. 
CBS = \frac{\text{Br ingested} - 10\%}{[\text{Br}]_e}

\text{Total water (TW) (}^2\text{H}_2\text{O space)} = \frac{\text{grams of } ^2\text{H}_2\text{O given}}{^2\text{H}_2\text{O conc (ppm) in plasma water}}

\text{TW} - \text{CBS} = \text{ICW (intracellular water)}

As an index of sexual maturation, DHEAS was measured in a small sample of plasma from several settlements by the methods described (13).

Ethical approval for this work was given by the chairman of the aboriginal councils and by the Department of Health, Western Australia.

STATISTICAL APPROACHES

Urine Analysis

For age, weight, and height, the means for the aboriginal and Caucasian groups were compared using \( t \)-tests. Using simple linear regression, there was no evidence in either group of any relationship between the zinc/creatinine ratios and age or height. Therefore the mean values for the two groups were compared using a \( t \)-test.

Body Composition

Regressions of total body water on extracellular volume and of intracellular water on the cube of height were calculated for the aboriginal and Caucasian groups, and the regressions for the two groups compared.

RESULTS

Indices of Zn Status

Figure 2 summarizes results obtained for RBC Zn, lymphocyte and granulocyte Zn, plasma Zn, and hair Zn compared with corresponding determinations in Caucasian schoolchildren from Adelaide. RBC zinc was significantly reduced for aboriginal children. The Zn per gram of hemoglobin was considered versus age in children 6 to 13 years old. Similarly, the Zn contents of lymphocytes and granulocytes were significantly reduced. As mentioned, a reduced level of Zn in hair has been a consistent observation. Plasma Zn was also reduced. Also of interest has been the constant occurrence of hypercupremia. For the La Grange settlement, 90% of the boys showed an elevation greater than 2 SD above the norm for 1981, 1982, and 1983. At the same time, approximately 50% of the boys and girls have shown hypoferremia at all settlements over the 9 years of investigation. La Grange is no exception.
FIG. 2. Indices of zinc status are considered for one settlement, "La Grange"(c). By comparison with Caucasian children (©) 5 to 13 years of age, values for RBC and WBC Zn content were reduced, as were values for plasma Zn and hair Zn: (A) RBC, µg Zn/g Hb, p<0.025; (B) plasma, µg Zn/dl, p<0.0005; (C) lymphocytes, ng Zn/10^7 cells, p<0.01; (D) polymorphs, ng Zn/10^7 cells, p<0.025; (E) hair, µg Zn/g hair, 50%<105 µg/mg.

Figure 3 depicts the urinary Zn/creatinine ratio for normal Caucasian children (11). The data for normal children were obtained in 1968, when methods for Zn analyses were less precise. The standard deviation and regression line for the ratio versus body length is shown. The open triangles reveal the points for aboriginal boys. Two-thirds of the points fall below 1 SD. Both sets of children were on a red-meat-free diet at the time of urine collection. Comparison of the ratio for aboriginal boys with Caucasian children studied in 1983 revealed significant differences (p<0.01) when mean values were compared. However, no relationship existed with Zn/creatinine and height.

Growth and Body Composition

Figures 4, 5, and 6 contain information with respect to anthropometric growth for 1981 to 1983. Points for all boys have not been plotted—only a sizable sample (20%). However, it is clear that weight is affected more than height-for-age, and that weight-for-stature also reveals reduced growth over the 3-year period. Growth retardation persists.

The relationship between extracellular volume (ECV) and total body water (TBW) is shown in Fig. 7 for normal Caucasian boys. The standard deviation is shown by the broken lines. The relationship for the normal child is very constant, and the line for Caucasian girls is identical (but not shown). Approximately 40% of body
Urinary excretion of Zn per unit creatinine is shown for aboriginal boys and compared with Caucasian children studied in 1968 (points not shown). The open triangles represent studies done in 1982. These studies including Caucasians were done when a red-meat-free diet was being consumed: Zn/creatinine (μg/mg) = 1.583 - 0.007(Ht); N = 33; r = -0.58.
FIG. 4. Body weights for age for aboriginal boys from La Grange settlement are plotted on charts provided by the National Center for Health Statistics from the United States. Each boy has been followed for 3 years (1981–1983); to simplify the figure only a few of the group of boys are considered. Growth retardation is persistent.

Water is extracellular and 60% intracellular during normal growth during childhood. By contrast, more than half of the points for aboriginal boys fall below 1 SD. When the two regression lines were compared, the line for data from aboriginal boys intersected the y-axis at a lower point (p<0.05). In other words, more fluid resides outside the body cells in aboriginal boys.

The relationship between intracellular water (an index of cell mass) and body length is described in Fig. 8. The relationship between intracellular water (ICW) and body length is a quadratic equation (14); however, if body length is cubed (2), one is considering volume against volume. Figure 8 shows that there is less cell mass for body length in aboriginal boys. The regression line for aboriginal boys again intercepts the y-axis (ordinate) at a lower point (p<0.01).

Eleven of the boys studied were not bled 3 hr after administration of $^2$H$_2$O but, rather, at 22 to 24 hr. Ordinarily, the losses of deuterium in urine and sweat would be minimal over this time. Of those bled at 3 hr (39 boys), the TBW was 646 ± 43 g/kg. For those bled at 22 to 24 hr, the TBW was 755 ± 43 g/kg (p<0.001). The relatively rapid fall in $^2$H$_2$O in the plasma supports the existence of an increased water turnover.
Sexual Maturation as Assessed by DHEAS

Table 1 contains information concerning plasma DHEAS levels. The 57 Caucasian children studied supplied data to document the normal situation. With height on the ordinate and DHEAS on the abscissa, an exponential curve (curving upward) was described. The data from each of the settlements (La Grange, One Arm Point, Lombadina, and Beagle Bay) were then considered, and separate curves for the data of children from each settlement were constructed. Table 1 contains predicted values for body lengths of 100 to 160 cm. When the data for DHEAS, as estimated by the fitted curves, are tabulated at discrete heights, the children from One Arm Point, Lombadina, and Beagle Bay show a markedly lower level of DHEAS in comparison to Caucasian children. Strangely, La Grange children did not show this difference. As age progressed toward puberty, the difference between aboriginal and Caucasian children became less. The significance of these findings ranged from $p<0.01$ to $p<0.005$ for the three settlements (Lombadina, Beagle Bay, and One Arm Point).
ZINC STATUS, COMPOSITION, AND MATURATION

FIG. 6. Similar to Fig. 4 except that weight versus stature is considered.

DISCUSSION

Zinc and the Middle East

In the earliest descriptions of zinc deficiency on the human, growth retardation and delayed sexual maturation were emphasized. Children were found to have hepatosplenomegaly, and anorexia was noted. Even though modern instrumentation was not available, reduced plasma zinc and iron levels were demonstrated together with hypercupremia (15).

With the passage of years, it appears that hepatosplenomegaly is not a feature, delayed sexual maturation is not always remarkable, and anorexia may or may not be present. The conclusion that in Iran the consumption of unleavened bread (almost to the exclusion of other foods) was responsible for zinc deficiency has found strong support because phytate renders the zinc unavailable. Perhaps the conclusion that intestinal parasites such as hookworms were responsible for the zinc deficiency in Egypt finds less support, since schistosomiasis is widespread throughout the world. Moreover, the consumption of unleavened bread is just as customary in Egypt as it is in Iran.

Studies of aboriginal children may clarify the true picture of the zinc-deficient state, which was difficult for workers in Iran and Egypt because of the presence
of other factors. Admittedly, geophagia, parasitic infections, and the consumption of purified flour is a finding among the aboriginal people, as indeed is the tenfold higher incidence of diabetes mellitus seen in adults (5).

More than 20 years later, it is now recognized that the widespread occurrence of hypozincemia, together with reduced Zn levels in hair in a population, is strong evidence that a zinc problem exists in a population. Walravens et al. (16), however, warned that such indices yield no information regarding the degree of zinc deficiency in the individual. They did find, however, that Mexican children of a poor socioeconomic background with (a) hypozincemia, (b) hair zinc reduced below 105 μg/g, or (c) evidence of a poor zinc intake do respond in growth to a 10-mg zinc supplement given over a 1-year period. Those with the poorest growth initially show the best growth responses.

FIG. 7. For normal children during growth, a strong linear relationship holds between body water and extracellular volume. The line for Caucasian boys is shown together with the SD (dotted lines). The open triangles represent points for aboriginal boys. Note that aboriginal boys have more fluid in the extracellular space.
FIG. 8. The volume of intracellular water (ICW) reflects cell mass and is plotted against the cube of height for aboriginal boys (open triangles). The regression line is shown. The regression line for Caucasian boys of the same age is recorded also, but no points are shown. The reduction of ICW relating to height cubed was significant ($p<0.01$).

Indices of Zinc Deficiency

Prasad and Cossack (17) have emphasized that the study of white blood cells (WBCs) in particular neutrophils, is more likely to indicate the true state of the cellular phase with respect to zinc than other indices. Mild zinc deficiency leads to changes in neutrophils after only 4 weeks, whereas plasma zinc levels do not change
TABLE 1. Mean predicted DHEAS levels at various heights based on exponential curve-fitting*

<table>
<thead>
<tr>
<th>Children 5–13 years</th>
<th>DHEAS (μmol/liter) at heights 100–160 cm</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>100</td>
</tr>
<tr>
<td>Adelaide (controls)</td>
<td>57</td>
</tr>
<tr>
<td>La Grange</td>
<td>80</td>
</tr>
<tr>
<td>One Arm Point</td>
<td>60</td>
</tr>
<tr>
<td>Lombadina</td>
<td>29</td>
</tr>
<tr>
<td>Beagle Bay</td>
<td>23</td>
</tr>
</tbody>
</table>

*The plasma DHEAS levels of Caucasian Adelaide children are compared with the values obtained at four settlements in the Kimberley (La Grange, Beagle Bay, Lombadina, and One Arm Point). The comparison has been made by comparing curves (see text) so that at any given height the value for DHEAS can be predicted for children in the settlement.

and 11 weeks pass by before RBCs show reduced levels on a truly zinc-deficient diet (18).

Unfortunately, at this time only a small amount of information is available in the literature of WBC zinc (19–27), and only a few investigators have separated lymphocytes from granulocytes (19) or monocytes from granulocytes (23) (Table 2). No information on children is available except for a few observations on 11 children by Nishi in Japan (22). Separation of WBCs into types seems important, since monocytes contain more Zn than granulocytes (23) and eosinophils are known to contain about 10 times the amount of zinc as other granulocytes. There is Zn binding to special peptides rich in arginine, as shown by Olsson et al. (28). Moreover, it is extremely difficult to be entirely sure that reagents are absolutely free of zinc. Clearly, after separation of cells, minimum handling prior to analysis is preferable. To this end we released only a few (but known) number of cells onto a 3-mm-diameter area of Mylar foil mounted on a 15-mm disc. We subjected these cells to proton-induced x-ray emission (PIXE). The use of PIXE for a single layer of cells seems to be encouraging, since in preliminary work there was not even an overlap for Zn levels between aboriginal WBCs and those from Caucasian children.

Whether urine and the Zn/creatinine ratio is a valuable index of low Zn status is difficult to judge, since little information on urinary excretion is available for children (11). It appears that urine is no more helpful than hair or plasma. There is reason to believe that the children studied in the La Grange settlement are zinc-deficient, since we have studied multiple indices of Zn status.

Growth and Body Composition

Persistent growth retardation during an extended period of reduced zinc status has never been shown before, and on the assumption that these children are significantly depleted, the documentation of their anthropometric charts is impor-
TABLE 2. Review of literature concerning zinc content of lymphocytes and granulocytes in normal individuals

<table>
<thead>
<tr>
<th>Subjects</th>
<th>Cell no.</th>
<th>Zinc (ng/mg or 10⁷ cells) (mean ± SD)</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Granulocytes</td>
<td>Lymphocytes</td>
<td>Both leukocytes</td>
</tr>
<tr>
<td>Children</td>
<td>10⁷</td>
<td>101 ± 29</td>
<td>22</td>
</tr>
<tr>
<td>Adults</td>
<td>10⁷</td>
<td>129 ± 33</td>
<td>22</td>
</tr>
<tr>
<td>Adults</td>
<td>10⁷</td>
<td>70 ± 20</td>
<td>23</td>
</tr>
<tr>
<td>Infants</td>
<td>mg•</td>
<td>51 ± 1.1 (SEM)</td>
<td>26</td>
</tr>
<tr>
<td>Pregnant mothers</td>
<td>mg•</td>
<td>58 ± 15 (SEM)</td>
<td>26</td>
</tr>
<tr>
<td>Adults</td>
<td>mg•</td>
<td>72 ± 16</td>
<td>20</td>
</tr>
<tr>
<td>Adults</td>
<td>10⁷</td>
<td>81 ± 10</td>
<td>20</td>
</tr>
<tr>
<td>Adults</td>
<td>10⁷</td>
<td>102 ± 13</td>
<td>27</td>
</tr>
<tr>
<td>Adults</td>
<td>10⁷</td>
<td>108 ± 19</td>
<td>24</td>
</tr>
<tr>
<td>Children</td>
<td>10⁷</td>
<td>95 ± 44</td>
<td>25</td>
</tr>
<tr>
<td>Children</td>
<td>10⁷</td>
<td>81 ± 26</td>
<td>3</td>
</tr>
<tr>
<td>Adults</td>
<td>10⁷</td>
<td>90 ± 1.7</td>
<td>19</td>
</tr>
<tr>
<td>Adults</td>
<td>10⁷</td>
<td>75 ± 20</td>
<td>40</td>
</tr>
</tbody>
</table>

*Dried protein 1 mg = (approx.) 10⁷ cells.

Important. It should be pointed out that in Queensland, Dugdale (29) found that at least in one settlement with good sanitation and superior food supplies, children grow such that on the anthropometric scale they follow the 50th percentile. This indicates that a genetic basis is not present. MacFarlane (30) studied the aboriginal desert people and found that water turnover was 190 ml/kg/day, compared with 80 ml/kg/day for Caucasians. He found increased extracellular volume and excessive sweating at a given but hot environmental temperature. The fall in ²H₂O concentration revealed here over a 1-day period supports the conclusion that water turnover is increased. The finding of a reduced cell mass raises the question of protein reserves (2,14) and whether protein synthesis is decreased or protein degradation increased. One might well speculate that nitrogen turnover is increased. Indeed, our own work 15 years ago revealed that in liver tissue under widely varying hormonal and nutritional situations, the RNA/DNA ratio was a mirror image of the Zn/DNA ratio (31).

The finding of expansion of the extracellular volume again supports the original arguments of MacFarlane and raises the question of whether this added extracellular volume protects the individual from desert heat and explains the prolonged survival of the aboriginal relative to the short-term demise of the Caucasian. Indeed, one could project such thinking and suggest that the reduced cell mass reduces body metabolic mass and thus increases endurance to heat. If zinc deficiency was selected as a means of protection against the environment, one would also have to argue that such a sacrifice would concomitantly ensure a loss of cell-mediated immunity (32). From a more realistic viewpoint, it can be noted that the higher-than-expected body water content of the aboriginal boys predicts reduced body fat, which is in line with their general appearance of a lean body with "matchstick" legs.
Sexual Maturation

Little attention has been given to the process of sexual maturation in the aboriginal. It is known that some aboriginal girls appear to mature early and may become pregnant as early as 13 years of age. On the other hand, zinc deficiency appears to affect the sexuality of boys more than girls. Brown (33) documented delayed bone maturation in aboriginal children. The present finding of reduced plasma levels of DHEAS, which is the precursor of the sex hormones (testosterone and estrogen) (34), in the neighboring settlements is a step forward in demonstrating sexual retardation. We cannot explain why the levels for La Grange children were normal. It must be acknowledged that any restriction in dietary intake (calories) leads to delayed maturation (35).

A Note on Hypercupremia

As already mentioned, the prevalence of an elevated plasma Cu concentration is far more obvious than the occurrence of hypozincemia. Several investigators have described hypercupremia as a genetic difference in different people (36,37). On the other hand, there does seem to be some inverse relationship between Zn and Cu, and administration of zinc in large amounts reduces levels of Cu.

Certainly, one cannot consider an elevated plasma Cu concentration as evidence of protein-energy malnutrition (PEM). Indeed, the early work of Holtzman et al. (38) drew attention to reduced levels during PEM. In fact, Castillo-Duran et al. (39) recommended the use of Cu supplements during PEM.

CONCLUSIONS AND FUTURE IMPLICATIONS

The studies conducted over the past 9 years have been protracted owing to costs, distance, and inaccessibility of the settlements. Initially, the work resembled a survey—the monitoring of plasma Zn, Cu, and Fe concentrations together with anthropometric measurements and measurements of cell mass. The entrance of the Commonwealth Scientific and Industrial Research Organisation (C.S.I.R.O.) under the direction of R. M. Smith led to confirmation of the plasma and anthropometric findings and drew attention to reduced hair zinc levels. Recently, the C.S.I.R.O. with our group initiated a full supplementary but double-blind study of 300 children 5 to 13 years of age. The present work allows inspection of multiple indices of Zn status. Further work during the period of supplementation and afterward should support the contention of Prasad and Cossack that WBCs are the true index of zinc deficiency or sufficiency, a thesis also elaborated by Hinks et al. in their studies of the effects of contraception in women (40). Presumably, the WBCs should reflect whether supplementary Zn is or is not effective, as indeed one might also judge from the rate of growth of the children.

Dietary intake has not yet been studied appropriately in these children, nor has the route of zinc loss. Casual inspection suggests that dietary intake is not a problem but repeated bowel infection may be. It is known that diarrhea enhances Zn losses
very significantly and reduces plasma insulin concentrations (41). Poor sanitation is a feature of missions or settlements, and viral, bacterial, and parasitic infestations are frequent in these people, who originally were nomadic. Thus attention to sanitation was never a requirement. On the other hand, geophagia is practiced and, as shown by Cavdar et al. (42), only 5 g of clay when ingested inhibits the absorption of zinc and iron.

The fact that sweat volume is increased by comparison with Caucasians also represents a potential source of Zn loss and raises the question of whether the problem discussed in this chapter extends below the Tropic of Capricorn.

In retrospect, the study of blood cells, urine, and hair all support the thesis that a Zn-deficient state exists. Growth retardation is persistent, sexual maturation as assessed by DHEAS is delayed, cell mass is reduced, and N turnover is probably increased. A case for Zn deficiency is strong, and supplementation should further clarify the situation.

The concept of adaptation cannot be totally rejected. These people represent the oldest race in the world. Over the 50,000-year period a low zinc status could have been selected as a measure of survival against desert heat, with increased water turnover, lower metabolic mass, and a reduced growth rate. One might speculate that they developed an alternative method to regulate cell-mediated immunity. However, as stated, Dugdale (29) found that under good socioeconomic conditions, the children grow along the fiftieth percentile. Cockington (43) made the same observation. Clearly, detailed studies in a sophisticated medical center are indicated. In the meantime, the implications for government are serious, and any plan for health programs should be viewed in the light of zinc status.

ACKNOWLEDGMENTS

This work was supported by The Channel 10 Children's Medical Research Foundation of South Australia. We would like to thank the Australian Associated Brewers for their support in the organization of a meeting in Perth to discuss this work. The help and collaboration of Dr. R. M. Smith is acknowledged, as is the increasing help from the Commonwealth Scientific and Industrial Research Organisation of Australia.

REFERENCES

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DISCUSSION

Dr. Prasad: You did not mention anything about the immunological aspect of your patients. Do you have any information regarding this?

Dr. Cheek: We only have information on aboriginal children with PEM gathered at a time when it was not appreciated that they had a zinc problem (Jose et al. Med J Aust 1975;2:699–705). Primary antigen recognition and blastogenic response of T-lymphocytes were significantly impaired. There was some increase of B- and T-lymphocytes, a normal secondary antibody response to tetanus toxoid, and some impairment of neutrophil chemotaxis with impairment of the metabolic response to phagocytic and intraphagocytic bactericidal activity. One cannot conclude that these findings are necessarily related to zinc deficiency. Indeed, it is difficult to differentiate immune changes due to PEM from those due to zinc (Goode et al. Zn and immunity. In: Prasad A, ed. Current topics in nutrition. New York: Alan R. Liss, 1982:189).

Dr. Golden: We are eagerly awaiting the supplementation results because it is the only way we are going to tell whether it is the adaptation or the malnutrition that is affecting the metabolism of zinc or zinc is affecting the metabolism of the patient. Aboriginals are metabolically very different from Caucasians in terms of most physiological measurements that have been made—their response to heat stress, their ability to withstand cold, and their ability to fast for very long periods of time with relatively few changes in body composition. Do you think that it is reasonable to compare aboriginal subjects with Caucasian normals? Are these aboriginals normal?

Dr. Cheek: That is an excellent question. In fact, I have given Dr. Alan Dugdale in Brisbane notice that at some stage we shall seek to study a group of children in a settlement in Queensland, where they have good nutrition, good sanitation, and grow along the 50th
centile, instead of the 5th. I do feel your comment is correct. These people have lived in one continent for 50,000 years. There are few other people on this planet that can make an equal claim. At the same time, I would ask the question: Has nature seen fit to reduce their zinc status? To reduce their cytoplasmic growth? To reduce their BMR? To render them more adaptable to heat? To provide less food at the expense of providing less energy for growth? Admittedly, with loss of cell mass their extracellular volume increase provides for dehydration. Clearly, the restudy of body composition with supplementation may provide some answers. I do not believe that a physiological loss of zinc occurs; if deficiency exists, it is pathological.

Dr. Golden: They have a different body composition. We have recently done a study on Blacks measuring skinfold thicknesses throughout pregnancy. We found that relative to Caucasian normals they were not increasing the skinfold of their arms, but at the superiliac and subscapular sites they were putting on enormous amounts of fat. Thus the accumulation of fat during pregnancy was quite different in Black and Caucasian women. The difference seemed to be genetic, as it was consistent through all the social classes. We must not lose sight of these genetic differences: 50,000 years is a long time to have spindly legs if it is abnormal.

Dr. Cheek: Genetic differences are of interest. This is why I have dwelled on the hypercupremia. There are several reports in the literature suggesting that elevation of plasma copper concentration in some individuals is genetic. The most recent (Kenny, Nutr Rep Int, 1983;27:1227–30) suggests such for the Indians of North America. The elevation of plasma copper is regarded by my colleague, Richard Smith, as genetic. I am not sure at this time, as there seems to be an inverse relationship in the body between zinc and copper. Robert Kirk has drawn attention to the fact that there are various types of a zinc-dependent enzyme, carbonic anhydrase, in RBCs of aboriginal people, a genetic finding (Med J Aust 1978;1:183–93).

Dr. Chandra: You implied that the changes in copper concentration may have been due to altered zinc intake. This is most unlikely. Secondly, you mentioned that the aboriginal children have low weight-for-height parameters and some evidence of zinc deficiency, and you suggested that one may be the cause of the other. Until you obtain the results of your supplementation trials, it would be difficult to conclude a causal relationship.

The clinical pattern of aboriginal subjects described by Dr. David Jose was different from what we recognize as obvious zinc deficiency. Even among the various aboriginal settlements in Australia, there may be very significant differences in deficiency patterns, perhaps related to the geochemical differences of various locations.

Dr. Cheek: Dr. David Jose's work was done a decade ago. I agree that the immune function should be studied, but Professor David Shearman in Adelaide believes that the patient has to be in the laboratory, not thousands of miles away. Even 6 hr delay with blood destroys the function of the WBCs. We know that weight-for-height is reduced in the aboriginal children (Aust NZ J Med 1981;11:508–12).

Dr. Gebre Mehedin: This may be irrelevant, but we have an increasing impression that a tendency toward hypercupremia is usually associated with marginal iron status.

Dr. Golden: Michael Gracey published a paper in Lancet last year of growth of fully breast-fed white children in Perth. The very low rates of weight gain in relationship to the data produced in the United States surprised many of us. Is there something peculiar about Western Australia, the Whites as well as the Blacks?

Dr. Cheek: The black babies are 400 g less in birth weight than the whites, as Professor J. Martin finds in Perth. Aboriginals living in the periphery of Perth are living under slum
conditions and do not do well, as Gracey has found. The children do better in settlements, but in settlements there is poor sanitation. Indeed, Dr. Geoff Davidson, gastroenterologist at The Adelaide Children's Hospital, is on the receiving end of black infants from Alice Springs with gastroenteritis. At times, he says it is difficult to maintain plasma zinc levels. He is impressed with the flattened epithelial cells in the intestine—the poor growth of the villi and the retarded cell multiplication at the crypt level. We are investigating stool zinc losses. Workers in Toronto have shown excessive zinc losses in gastroenteritis.

**Dr. Prasad:** I would like to say one thing about the skin lesions in Egyptian and Iranian dwarfs. It did not resemble acrodermatitis enteropathica lesions. All I saw was some roughening of the skin in the face and exposed parts of the body. My first impression was that it was due to vitamin A deficiency except that the vitamin A level was normal. My other comment is with respect to the copper/zinc ratio. In my experience high copper is better correlated with low zinc. In malignancies, leukemias, and lymphomas, a high copper/zinc ratio is regarded as an indicator of activity of the malignancy. In our studies with the zinc-deficient human model we have observed that plasma copper goes up. Also in patients with renal disease and sickle cell disease the plasma copper/zinc ratio is high. Thus I believe that a high plasma copper level correlates reciprocally with low plasma zinc rather than with low plasma iron.

**Dr. Chandra:** We generally expect perturbation of vitamin levels in zinc deficiency. Dr. Prasad, what is the explanation for normal levels in the face of obvious zinc problems in your patients?

**Dr. Prasad:** This is not a very definite or consistent relationship. In our experimental zinc-deficient human model, I never saw any change in serum vitamin A levels as a result of dietary zinc restriction. It could be that we were dealing with a mild variety of zinc deficiency.

**Dr. Golden:** If it is only in moderately severe deficiency, the suggestion which has been made—that one could use the dark adaptation test as a test of subtle zinc deficiency—should not be valid.

**Dr. Prasad:** Now the story is a bit different. There is an enzyme, retinene reductase, which is a zinc-dependent enzyme. This enzyme is responsible for the conversion of retinol to retinene. We have suggested that abnormal dark adaptation in zinc deficiency is related to decreased activity of the zinc-dependent enzyme retinene reductase. Thus one might see abnormal dark adaptation in zinc deficiency despite normal levels of retinol-binding protein and vitamin A in the plasma.