

Micronutrients and its Correlation With Mental Performance Among Schoolchildren in Bario, Sarawak: A Preliminary Study

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Summary

Children who are born in a community with insufficient micronutrients, particularly iodine in remote rural areas are associated with low intellectual functions and mental retardation. The aim of this cross-sectional study is to determine the mental performance of the schoolchildren in Bario, a highland settlement of the Kelabit people in the district of Baram, Sarawak and to determine its correlation with the availability of iodine in the environment, as well as other micronutrients such as selenium, copper and manganese. A total of 25 schoolchildren in Bario age ranging from 7 to 12 years old participated in the study. Mental performance of the schoolchildren were tested using TONI-2 (Test of Nonverbal Intelligence - second edition), a cognitive ability measures with a response format which eliminates language and reduces motoric and cultural factors. The iodine levels from several wells, soils and salt found in Bario were determined using HPLC (software version 3.05.01) whilst serum levels of selenium, copper and manganese were measured using Graphite Furnace Atomic Absorption Spectrophotometer (GFAAS). The results showed that the median and mode scores of intelligence quotient for Bario were 82 and 75, respectively, whilst median and mode scores of intelligence percentile were 11.0 and 5.0, respectively. The maximum score achieved were at the 'average' level with the quotient score between 90 - 110. It was found that salt produced from one of the wells in Bario contained high quantity of iodine. Based on standards established by the Trace Elements Laboratory, Roben Institute, University of Surrey, United Kingdom, schoolchildren in Bario are having sufficient blood levels of copper and a high selenium and manganese levels. Despite the remoteness of the study area, the schoolchildren in Bario, Sarawak showed higher mental performance compared to other isolated areas. This is probably correlated with the high micronutrients availability, particularly iodine, found naturally in Bario.

Key Words: Micronutrient, Mental performance, Schoolchildren, Remote rural, Bario

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Introduction

A lot of micronutrients have beneficial effects on human health. They involved in many biochemical and physiological processes necessary for optimum growth, development and health; for example, iodine promotes normal function of the thyroid gland and prevents goitre, copper promotes normal red blood cell formation and protects against cardiovascular disease, selenium acts as antioxidant and protects against increased oxidation, and manganese promotes normal growth and development and aids in carbohydrate metabolism¹.

Iodine has an important role in growth and brain development. Iodine deficiency can cause either maternal and/or fetal hypothyroidism². Maternal hypothyroxinemia is a characteristic frequently seen in goitre endemic areas³, and low levels of maternal thyroxine (T4) has been shown to be subsequently correlated with the offspring intellectual products⁴. The recovery of maternal T4 to normal levels through iodine prophylaxis before pregnancy or at the first trimester will prevent endemic cretinism and subclinical deficit of intellectual and motor functions⁵. Trowbridge (1972)⁶ and Querido (1975)⁷ showed the evidence of intellectual capacity reduction in all individuals born in iodine-deficient community. Neurological damage due to iodine deficiency is not only limited to the syndrome of endemic cretinism, it may also show the effect of milder disorder, seen as the general reduction of intellectual potential in a community⁸. Children who were born in an iodine-deficient community particularly in remote rural areas are frequently associated with the reduction in intellectual functions and mental retardation.

Copper functions as a component in numerous metalloenzymes acting as oxidases to achieve the reduction of molecular oxygen⁹. It is also known as a mineral that promotes normal red blood cell formation, assists in production of several enzymes involved in respiration, promotes connective tissue formation and central nervous system function, and is part of superoxide dismutase and its antioxidant capacity¹.

Selenium is largely obtained from bread and cereals, fish, poultry, and meat. It plays a vital part in many metabolic functions¹⁰. Selenium is a key component of a number of functional selenoproteins required for normal health. The best known of these are the antioxidant glutathione peroxidase enzymes, which remove hydrogen peroxide and damaging lipid and phospholipid hydroperoxides generated in vivo by free radicals and other oxygen derived species. Selenium also plays an important role in the control of thyroid hormone metabolism. The iodothyronine deiodinases, which are responsible for the conversion of T4 to its active form, T3, are selenoenzymes¹¹. Selenium deficiency may cause reduced growth rates owing to a feedback response which lowers T3 mediated synthesis of growth hormone in the pituitary¹², while a combined deficiency of selenium and iodine exacerbates hypothyroidism¹¹.

Manganese is concentrated in the cells of the pituitary gland, liver, pancreas, kidney and bone. It stimulates the production of cholesterol by the liver and is a co-factor in many enzymes. Manganese is also involved in anti-oxidation process. Manganese is abundant in many foods such as beans, bran, carrots, chestnuts, hazelnuts, oatmeal, peanuts, peas, pecans, seaweed, spinach, tea and whole grains. Manganese promotes normal growth and development, helps many enzymes generate energy, aids in carbohydrate metabolism, promotes nerve function, aids in formation of connective tissue and involved in antioxidation process. Deficiency symptoms include abnormal growth and development of children. Deficiencies are extremely rare because manganese is widely available in the food supply and requirements are very small¹.

The aim of this preliminary study is to determine the mental performance of the schoolchildren in Bario, a highland settlement of the Kelabit people in the district of Baram, Sarawak and its correlation with the availability of iodine in the environment, as well as other micronutrients. The community has been known to have many elitists and known state figures despite its remoteness and isolation.

Materials and Methods

Study area

Bario, or the meaning in local term 'strong wind', is a highland remote rural area. It is located at approximately 3,500 feet above sea level. Bario is surrounded by hills and mountains (Basin shape) and the average temperature during dry season is 17°C. The number of children between 7 - 14 years old in this area were 195, whilst the average number of person per family were 3.4. There were approximately 248 families in Bario. The mode of transportation in this area were by road, walking and boat, whilst travel between districts were by air. The majority of the villagers (78.4%) were from Kelabit ethnic group and nearly all of them were Christian (Evangelist). Most of the settlers here (93.4%) were farmers.

Study subjects and sampling

A total of 25 schoolchildren in Bario aged 7 - 12 years old participated in this cross-sectional study. The subjects were selected using simple random sampling (Fisher random numbers) from the school register.

Mental performance

Mental performance of the schoolchildren were tested using TONI-2 (Test of Nonverbal Intelligence - second edition), a cognitive ability measure with a response format which eliminates language and reduces motoric and cultural factors. The test consisted of 55 abstract/figural problem solving. It is intended to be used with subjects aged 5 - 85 years. There are two equivalent forms of TONI-2, namely form A and form B, each containing 55 items arranged in order of difficulty. Since the test is administered for the first time in this group, Form A was used (Form B will only be used if there were subsequent occasions). Total raw score of the subjects were calculated and then converted to two kinds of normative scores: percentile ranks and deviation quotients¹³. Percentile ranks indicate the percentage of scores in the normative group that are above or below the

score in question, whilst Table I provides some useful guidelines for interpreting quotients.

Iodine levels in water and soil

The iodine levels of water and soil in selected locations were measured using HPLC (Software Version 3.05.01). A sample of man-made salt produced from the selected wells was also measured.

Serum levels of copper, selenium and manganese

Venous blood samples were drawn for serum copper, selenium and manganese analysis. Samples were taken without anticoagulant into Vacutainer tubes and then centrifuged at room temperature for 15 minutes at 1,500 G. The serum were then transferred to vials and stored at -20°C. The samples were then analyzed using Graphite Furnace Atomic Absorption Spectrophotometer (GFAAS). All glasswares used for analysis were washed with 10% nitric acid and rinsed with deionised water.

Clinical evaluation

The children's weight, percent body fat, fat mass, lean body mass and total body water were measured using TANITA Bodyfat Analyzer (model TBF-105, Tanita Corporation, Japan). Height without shoes was measured by the Microtoise tool (Depose, France), whilst blood pressure and pulse rate were measured using a digital blood pressure/pulse monitor set (model HEM-400C, Omron Corporation, Japan) which was taken three times.

Statistical analysis

The software for statistical analysis was the SPSS package (Statistical Package for Social Science, Version 10.0). All values were given as mean \pm SD, unless otherwise stated. For statistical evaluation, analysis of variance (ANOVA), t-test and chi-square were used to observe the difference between

groups or variables. In the linear regression analysis, the males was referred as code 1, whilst the females was referred as code 2. A probability of 5% or less was considered significant.

Results

Demographic profiles

The demographic characteristics of study subjects are listed in Table II. No significant different was observed between males and females in all characteristics studied.

Mental performance

Among the schoolchildren of Bario, the males showed higher median and mode scores in both percentile ranks and deviation quotients as compared to the females (Table III). As in the quotient distribution, the highest score achieved was the 'average' score (90 - 110) (Table IV). Majority of the male schoolchildren (63.64%) scored the highest score. As for the females, majority of them showed an equal distribution between the 'poor' (30.77%) and 'very poor' (30.77%) scores. Overall, majority of the schoolchildren in Bario (41.67%) gained the 'average' score.

Iodine levels in salt

A very high level of iodine was found in the man-made salt obtained from the well water in Pa' Umor, Bario. The level obtained was 445.5135 ± 0.0191 ppm, which was more than 44 times higher compared to iodine levels in well water, river water and soil in the same area (Table V).

Iodine levels in the environment

The highest level of iodine in the environment was found in the well water in Pa' Umor (Table V). This is followed by the well water in Pa' Main, the soil of Padang Pasir padi field, the soil 1 metre away from Pa' Umor's well, the well water in Long Banga, the soil 1.5 metres away from Pa' Umor's well and Pa' Umor river water. Man-made salt derived from wells in Bario was shown to have high levels of iodine.

Serum copper, selenium and manganese levels among the schoolchildren

Based on standards established by the Trace Elements Laboratory, Roben Institute, University of Surrey, Guilford, Surrey, United Kingdom, mean levels of copper and selenium were within the normal range, whilst mean levels of manganese was higher than the normal (Table VI). Meanwhile, comparison between gender showed no significant difference in all the micronutrients studied.

Correlation between variables

From the correlation analysis, among the important association observed were between age and lean body mass ($r = 0.61$, $p < 0.01$), lean body mass and serum copper levels ($r = -0.43$, $p < 0.05$), and systolic blood pressure and serum manganese levels ($r = 0.52$, $p < 0.01$) (Table VII). The results indicated that lean body mass of the schoolchildren increased with age, serum copper levels of the schoolchildren reduced with the increase in lean body mass, and serum manganese levels of the schoolchildren increased with the increase in their systolic blood pressure.

Table I: Guidelines on how to interpret TONI-2 Quotients

QUOTIENT	I.Q. LEVELS
>130	Very Superior
121-130	Superior
111-120	Above Average
90-110	Average
80-89	Below Average
70-79	Poor
<70	Very Poor

Table II: Clinical characteristics of study subjects

Clinical characteristics	The schoolchildren of Bario, Sarawak		
	Male (n = 12)	Female (n = 13)	Total (n = 25)
1. Age (year)			
Mean	8.7 ± 1.48	9.23 ± 1.42	9.00 ± 1.44
Range	(7 - 11)	(7 - 11)	(7 - 11)
2. Weight (kg)			
Mean	23.38 ± 5.37	26.06 ± 5.96	24.77 ± 5.73
Range	(15.30 - 32.50)	(19.30 - 42.20)	(15.30 - 42.20)
3. Height (cm)			
Mean	120.73 ± 9.00	123.65 ± 7.26	122.24 ± 8.08
Range	(104.00 - 137.60)	(112.00 - 136.50)	(104.00 - 137.60)
4. Percent body fat (%)			
Mean	13.93 ± 2.82	16.22 ± 4.35	15.12 ± 3.80
Range	(7.60 - 17.60)	(11.70 - 28.40)	(7.60 - 28.40)
5. Fat mass (kg)			
Mean	3.30 ± 1.15	4.43 ± 2.49	3.89 ± 2.01
Range	(1.40 - 5.70)	(2.50 - 12.00)	(1.40 - 12.00)
6. Lean body mass (kg)			
Mean	20.08 ± 4.44	21.63 ± 3.66	20.88 ± 4.05
Range	(12.90 - 28.10)	(16.80 - 30.20)	(12.90 - 30.20)
7. Total body water (kg)			
Mean	14.68 ± 3.26	15.83 ± 2.68	15.28 ± 2.97
Range	(9.40 - 20.60)	(12.30 - 22.10)	(9.40 - 22.10)
8. Systolic blood pressure (mm Hg)			
Mean	97 ± 9	100 ± 9	98 ± 9
Range	(83 - 115)	(85 - 116)	(83 - 116)
9. Diastolic blood pressure (mm Hg)			
Mean	62 ± 9	61 ± 10	61 ± 9
Range	(45 - 74)	(49 - 79)	(45 - 79)
10. Pulse rate (per minute)			
Mean	91 ± 16	94 ± 12	93 ± 14
Range	(78 - 138)	(75 - 116)	(75 - 138)

Data are mean values ± SD.

Table III: Mental performance of the schoolchildren in Bario, Sarawak

Mental performance indices	The schoolchildren of Bario, Sarawak (Kelabit, n = 25)		
	Male (n = 11)	Female (n = 13)	Total
Quotient:			
Median	92	75	82
Mode	92	75	75
Range	69 - 108	57 - 106	57 - 108
Percentile:			
Median	30.0	5.0	11.0
Mode	30.0	5.0	5.0
Range	2.0 - 70.0	0.5 - 66.0	0.5 - 70.0

Table IV: Quotient distribution of the schoolchildren in Bario, Sarawak

Quotients distribution	The schoolchildren of Bario, Sarawak (Kelabit, n = 25)		
	Male (%)	Female (%)	Total (%)
>130- (very superior)	-	-	-
121 - 130 (superior)	-	-	-
111.- 120 (above average)	-	-	-
90 - 110 (average)	63.64 (7)	23.08 (3)	41.67 (10)
80 - 89 (below average)	27.27 (3)	15.38 (2)	20.83 (5)
70 - 79 (poor)	0 (0)	30.77 (4)	16.67 (4)
<70 (very poor)	9.09 (1)	30.77 (4)	20.83 (5)

Table V: Iodine levels in water and soils in selected locations in Bario, Sarawak

Samples	Iodine levels (µg/g)
1. Well water (natural source)	
Pa' Main	5.9350 ± 0.0495
Pa' Umor	8.0148 ± 0.9082
Long Banga	0.7620 ± 0.0170
2. River water	
Pa' Umor	0.7020 ± 0.0028
3. Soil	
Site A (soil 1 metre away from Pa' Umor well)	0.8535 ± 0.0050
Site B (soil 1.5 metre away from Pa' Umor well)	0.7415 ± 0.0021
Padang Pasir padi field	2.9340 ± 0.0481
ANOVA test	F = 226.07 p < 0.0001

Table VI: Mean serum levels of copper, selenium and manganese among the schoolchildren of Bario, Sarawak

Micronutrient/Standard	Mean ± sd			Surrey Standard
	Male (n=12)	Female (n=12)	Total (n=12)	
Copper (µmol/L)				
Mean	17.46 ± 5.09	16.21 ± 3.02	16.84 ± 4.14	11 - 20
Range	(11.16 - 28.41)	(10.99 - 21.62)	(10.99 - 28.41)	
Selenium (µmol/L)				
Mean	1.20 ± 0.35	1.12 ± 0.21	1.16 ± 0.29	0.57 - 0.90
Range	(0.67 - 2.06)	(0.74 - 1.46)	(0.67 - 2.06)	(2 - 15 years old)
Manganese (µg/L)				
Mean	1.34 ± 0.84	1.48 ± 0.71	1.41 ± 0.76	0.5 - 1.3
Range	(0.40 - 3.40)	(0.50 - 2.60)	(0.40 - 3.40)	

Table VII: Correlations between age, nutritional status, clinical status, mental performance and micronutrient levels among the schoolchildren in Bario Sarawak

Variables	Statistical values	BMI	% Body fat	LBM	Quotient	Percentile	SBP	DBP	PR
Age	r	0.22	0.01	0.61	0.20	0.13	0.18	-0.19	-0.15
	p	0.293	0.945	0.001**	0.347	0.546	0.400	0.360	0.480
Copper	r	-0.23	0.18	-0.43	-0.12	0.05	0.26	0.33	0.3
	p	0.287	0.406	0.035*	0.587	0.817	0.230	0.111	0.146
Selenium	r	-0.03	0.24	-0.18	0.21	0.03	0.01	-0.11	-0.16
	p	0.905	0.267	0.397	0.332	0.873	0.960	0.596	0.457
Manganese	r	0.17	0.32	0.09	0.29	0.28	0.52	0.09	-0.15
	p	0.422	0.134	0.686	0.176	0.180	0.009**	0.669	0.483

BMI - Body Mass Index; LBM - Lean Body Mass; SBP - Systolic Blood Pressure; DBP - Diastolic Blood Pressure; PR - Pulse Rate; r - correlation coefficient; p - probability; **p < 0.01; *p < 0.05; significant.

Discussion

The link between iodine deficiency during pregnancy and mental retardation in the offspring has been recognized for nearly 100 years¹⁴. The current study shows that sufficient levels of micronutrient, particularly iodine can positively affect the schoolchildrens' mental performance. Copper, selenium, manganese and iodine are considered as the nutritionally essential trace minerals where the intake should be adequate to prevent deficiency yet not enough to cause toxic effects¹⁵. One advantage of Bario as compared to other isolated areas studied in Malaysia was its high iodine content found naturally in its soil¹⁶⁻¹⁸. Bario has several number of wells containing high levels of salt. It is from these salts that high concentration of iodine was found. Therefore, the community in Bario, particularly the schoolchildren, are exposed to local natural food of high micronutrient content. This is strengthened through observations and interviews that none of the villagers in Bario have visible goitre. The schoolchildren in Bario showed better mental performance than the other isolated areas studied¹⁶⁻¹⁸. Therefore, it is predicted that sufficient intake of micronutrients among the schoolchildren in Bario might have improved their mental performance. However, further study need to be conducted to confirm these findings.

A study conducted by Fordyce et al. (2000)¹⁹ in Sri Lanka showed that iodine concentrations in shallow well water samples from the low goitre incidence area (prevalence of goitre below 10%) were 53 to 84 µg/L (0.053 to 0.084 µg/g), which were distinctly higher than those from the moderate goitre incidence area (prevalence of goitre 10 - 25%) and high goitre incidence area (prevalence of goitre above 25%) (3.3 - 23.5 µg/L or 0.0033 - 0.0235 µg/g). However, iodine concentrations in well and river waters in Bario showed approximately a 100 times higher as compared to the values in the low goitre incidence area in Sri Lanka. Based on the same study by Fordyce et al. (2000)¹⁹, total concentrations of iodine in soil for both goitre prevalent and non-

goitre prevalent villages in Sri Lanka ranged from 0.13 to 10 µg/g, and this value was comparable to values reported for soils in other regions of the world²⁰. The total concentrations of iodine found in soil of Bario (soil taken near Pa' Umor's well and at Padang Pasir padi field) ranged from 0.74 to 2.93 µg/g, which is a smaller range compared to the Sri Lanka's values. In this study, whilst water iodine concentrations can be described as very high in Bario, soil iodine concentrations can be explained as average to marginal when compared with water and soils from Sri Lanka and possibly other regions of the world.

When clinical characteristics of study subjects were compared with the *Orang Asli* schoolchildren in Legap Post, Perak of similar age group, it was found that the schoolchildren in Bario were significantly heavier, has higher percentage of body fat, higher fat mass and lower systolic blood pressure as compared to the *Orang Asli* schoolchildren^{17,18}. Better nutritional status among the Bario schoolchildren again could be due to better accessibility to selected micronutrients. This is supported by their mental performance in which they showed significantly higher quotient deviation (median value: 82 versus 67) and higher percentile rank (median value: 11 versus 1) scores as compared to the *Orang Asli* schoolchildren^{17,18}, and this is true in both gender. It was also noted that the overall mental performance of the schoolchildren in Bario was 'average', whilst the schoolchildren in Legap Post showed 'very poor' despite of the comparable remoteness of both areas^{17,18}.

The schoolchildren in Bario was also noted to have sufficient serum levels of several micronutrients studied, namely copper, selenium and manganese. Based on the range showed in the study, the schoolchildren were at the upper range of selenium and manganese levels. The mean serum selenium levels in this study was 1.16 ± 0.29 µmol/L, with the range of 0.67 to 2.06 µmol/L. This is comparable with a study in Italy where the serum selenium range values reported were 0.53 to 2.01 µmol/L²¹. Meanwhile, mean serum

selenium among adults living in seleniferous area were $2.10 \pm 0.38 \mu\text{mol/L}$ 22. Some studies showed that plasma selenium levels increase with age from neonate until adult (16 years old) before it get stabilizes^{23,24} but such correlation was not shown in the study. This study, however, showed some associations between age and lean body mass, copper levels and lean body mass, and manganese levels and systolic blood pressure. As the children grow, their lean body mass will increase, and their serum copper levels will decrease. When the levels of manganese increase, systolic blood pressure of the children will also increase. This might be related to one of the functions of manganese in the body, that is to stimulate the production of cholesterol by the liver. The reduction of copper levels, however, cannot be explained. Gender caused no significant different in serum selenium levels, as previously been observed in healthy subject^{25,26}. However, serum manganese levels was significantly higher in female compared to male subjects. Males absorbed significantly less manganese than females²⁷. This difference may be related to iron status. Effect on laboratory tests also showed that excess manganese can reduce serum iron¹. Study by Finley (1999)²⁸ showed that high ferritin

concentrations were associated with reduced manganese absorption. Serum ferritin concentrations were higher in males, therefore it would lower the bioavailability of manganese. Since iron status was not measured in this study, the bioavailability relationship between serum iron and manganese cannot be highlighted.

Conclusion

It can be concluded that apart from the remoteness of the study area, the schoolchildren in Bario, Sarawak have better mental performance than other isolated areas studied. This is probably correlated with the high micronutrient availability in the environment, particularly iodine found naturally in Bario.

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References

1. Griffith HW. Minerals, supplements & vitamins - the essential guide. Tucson, Arizona: Fisher Books, LLC, 2000.
2. Boyages SC, Halpern JP. Endemic cretinism: toward a unifying hypothesis. *Thyroid* 1993; 3: 59-69.
3. Delange F, Thilly C, Bourdoux P, et al. Influence of dietary goitrogens during pregnancy in humans on thyroid function of the newborn. In: Delange F, Ittekkk FB, Ermans AM, (eds). Nutritional factors involved in the goitrogenic action of cassava. Ottawa: International Development Research Centre, 1982; 40-50.
4. Pharoah POD, Connolly KJ, Ekens RP, Harding A. Maternal thyroid hormone levels in pregnancy and the subsequent cognitive and motor performance of the children. *Clin Endocrinol (Oxf)* 1984; 21: 265-70.
5. Pharoah POD, Connolly KJ. A controlled trial of iodinated oil for the prevention of endemic cretinism: a long-term follow-up. *International Journal of Epidemiology* 1987; 16(1): 68-73.

6. Trowbridge FL. Intellectual assessment in primitive societies, with a preliminary report of a study of the effects of early iodine supplementation on intelligence. *Advances in Experimental Medicine & Biology* 1972; 30: 137-49.
7. Querido A. Endemic cretinism - a continuous personal educational experience during 10 years. *Postgraduate Medical Journal* 1975; 51: 591-99.
8. Boyages SC, Collins JK, Maberly GF, Jupp JJ, Morris J, Eastman CJ. Iodine deficiency impairs intellectual and neuromotor development in apparently-normal persons. A study of rural inhabitants of north-central China. *Medical Journal of Australia* 1989; 150: 676-82.
9. Trumbo P, Yates AA, Schlicker S, Poos M. Dietary reference intakes: Vitamin A, vitamin K, arsenic, boron, chromium, copper, iodine, iron, manganese, molybdenum, nickel, silicon, vanadium, and zinc. *Journal of the American Dietetic Association* 2001; 101: 294-301.
10. Rayman MP. Dietary selenium: time to act. *British Medical Journal* 1997; 314: 387-91.
11. Arthur JR, Bermano G, Mitchell JH, Hesketh JE. Regulation of selenoprotein gene expression and thyroid hormone metabolism. *Biochem Soc Trans* 1996; 24: 384-88.
12. Arthur JR, Nicol F, Rae PWH, Beckett GJ. Effects of selenium deficiency on the thyroid gland and on plasma and pituitary thyrotrophin and growth hormone concentrations in the rat. *Clin Chem Enzymol Commun* 1990; 3: 209-14.
13. Brown L, Sherbenou RJ, Johnsen SK. Test of nonverbal intelligence: a language-free measure of cognitive ability. Second Edition Manual, Pro-Ed, 8700 Shoal Creek Boulevard, Austin, Texas, 1990.
14. Gardner LI. Historical notes on cretinism. In : Gardner LI, (ed). *Endocrine and genetic diseases of childhood and adolescence*. 2nd ed. Philadelphia: W.B. Saunders, 1975: 234-38.
15. Nutrition Committee of the Canadian Paediatric Society. Nutrient needs and feeding of premature infants. *Clinical Practice Guidelines*. *Can Med Assoc J* 1995; 52(11): 1765-85
16. Zaleha MI, Osman A, Iskandar ZA, et al. The supplementation of levothyroxine among indigenous people in endemic goitre areas: The impact of therapy. *Asia Pacific J Clin Nutr* 1998; 7 : 138-50.
17. Zaleha MI, Iskandar ZA, Khalid BAK, Osman A. Effect of iodized oil supplementation on thyroid hormone levels and mental performance among Orang Asli schoolchildren and pregnant mothers in an endemic goitre area in Peninsular Malaysia. *Asia Pacific J Clin Nutr* 2000; 9: 274-81.
18. Zaleha MI. The effect of levothyroxine and iodized oil supplementation in IDD prevention on thyroid hormone levels, nutritional status and mental performance among the aborigines in Malaysia. (PhD thesis), University Kebangsaan Malaysia, Kuala Lumpur, Malaysia, 1999.
19. Fordyce FM, Johnson CC, Navaratna URB, Appleton JD, Dissanayake CB. Selenium and iodine in soil, rice and drinking water in relation to endemic goitre in Sri Lanka. *The Science of the Total Environment* 2000; 263: 127-41.
20. Fordyce FM, Johnson CC, Navaratna URB, Appleton JD, Dissanayake CB. Studies of selenium geochemistry and distribution in relation to iodine deficiency disorders in Sri Lanka. *British Geological Survey Overseas Geology Series Technical Report WC/98/23*, 1998.
21. Lockitch G. Selenium: clinical significance and analytical concepts. *Crit Rev Clin Lab Sci* 1989; 27: 483-541.
22. Swanson CA, Longnecker MP, Veillon C, Howe M, Levander OA, Taylor PR, McAdam PA, Brown CC, Stampfer MJ, Willet WC. Selenium intake, age, gender, and smoking in relation to indices of selenium status of adults residing in a seleniferous area. *American Journal of Clinical Nutrition* 1990; 52(5): 858-62.
23. Wasowicz W, Zachara BA. Selenium concentrations in the blood and urine of a healthy Polish sub-population. *J Clin Chem Clin Biochem* 1987; 25: 409-12.
24. Navarro F, Navas P, Burgess JR, Bello RI, De Cabo R, Arroyo A, Villalba JM. Vitamin E and selenium deficiency induces expression of the ubiquinone-dependent antioxidant system at the plasma membrane. *FASEB Journal* 1998; 12(15): 1665-73.
25. Navarro M, Lopez H, Ruiz MI, Gonzalez S, Perez V, Lopez MC. Determination of selenium in serum by hydride generation atomic absorption spectrometry for calculation of daily dietary intake. *Sci Total Environ* 1995; 175: 245-52.
26. Gomez C, Ruiz-Lopez MD, Artacho R, Navarro M, Puertas A, Lopez MC. Serum selenium in

institutionalized elderly subjects and its relationship with other nutritional parameters. *Clin Chem* 1997; 43: 693-94.

27. Finley JW, Johnson PE, Johnson LK. Sex affects manganese absorption and retention by humans

from a diet adequate in manganese. *Am J Clin Nutr* 1994; 60: 949-55.

28. Finley JW. Manganese absorption and retention by young women is associated with serum ferritin concentration. *Am J Clin Nutr* 1999; 70: 37-43.