Effect of a six-month iron-zinc fortified milk supplementation on nutritional status, physical capacity and speed learning process in Indonesian underweight schoolchildren: randomized, placebo-controlled

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Abstrak

Tujuan *mengevaluasi efek suplementasi susu yang diperkaya dengan zat besi dan zink terhadap indikator pertumbuhan, kapasitas fisik dan kecepatan berpikir anak sekolah yang kurang berat (underweight).*

Metode *Eksperimen pada dua kelompok paralel tersamar ganda dilaksanakan di beberapa sekolah dasar di Jakarta dan Surakarta, Indonesia. Sejumlah 245 anak sekolah usia 7-9 tahun yang underweight di daerah miskin perkotaan Jakarta and Solo dialokasikan secara acak menerima dua gelas susu yang diperkaya zat besi dan zink (n = 121) atau susu biasa (n= 124) setiap hari selama enam bulan. Indikator biokimia, antropometri, kapasitas fisik dan fungsi kognisi diukur saat awal, bulan ke-3 dan ke-6.*

Hasil *Antara kelompok susu diperkaya dibandingkan dengan kelompok susu biasa, peningkatan kadar hemoglobin (berturut-turut 0,01±0,96 mg/dL dan 0.17 ± 0.81 mg/dL) dan serum feritin (berturut-turut 12,77 ± 25,50 mcg/dL dan 14,99 ± 29,56 mcg/dL) tidak berbeda bermakna antara kedua kelompok. Selanjutnya, terjadi penurunan kadar serum zink (berturut-turut 3,01 ± 3,24 mMol/dL dan 3,12 ± 3,71mMol/dL) yang tidak bermakna antara kedua kelompok. Penambahan berat badan pada kelompok susu diperkaya (1,31 ± 0,69 kg) berbeda secara bermakna (P=0.045) dibandingkan dengan kelompok susu biasa (1,13 ± 0,69 kg), sejalan dengan peningkatan indikator underweight (WAZ) pada kelompok susu diperkaya (1.47 ± 0.50) yang lebih besar secara bermakna (P=0.025) dibandingkan dengan peningkatannya pada kelompok susu biasa (1,33 ± 0,47). Terdapat perbaikan skor kecepatan berpikir yang bermakna (P=0.001) pada kelompok susu diperkaya (12,74 ± 11,76) dibandingkan dengan kelompok susu biasa (8.31 ± 9,60), namun tidak ada perbedaan yang bermakna untuk perbaikan skor kapasitas fisik antara kedua kelompok (berturut-turut 10323,77 ± 9253,83 dan 8435,94 ± 8824,55).*

Kesimpulan *pada anak sekolah usia 7-9 tahun yang underweight, suplementasi susu yang diperkaya dengan zat besi dan zink akan lebih mendukung pertumbuhan dan kecepatan berpikir.* ***(Med J Indones 2009; 18: 193-202)***

Abstract

Aim to evaluated the effect of milk supplementation enriched with iron and zinc on indicators of growth, physical capacity and cognitive performance in underweight school-children.

Method Two-armed, randomized controlled double-blind trial was performed in several primary schools in Jakarta and Surakarta, Indonesia. A total of 245 underweight schoolchildren aged 7-9 years living in urban poor areas of Jakarta and Solo were randomly allocated to receive two cups of iron-zinc fortified-milk (n = 121) or non-iron-zinc fortified milk (n= 124) supplementation daily for six months. Biochemical indicators, anthropometric indices, physical capacity and cognitive performance were measured at before and after the supplementation.

Results The study shows that between the fortified and non-fortified milk group, there was no significant different in haemoglobin increase (0.01 ± 0.96 mg/dL versus 0.17 ± 0.81 mg/dL) nor serum ferritin increase (12.77 ± 25.50 mcg/dL versus 14.99 ± 29.56 mcg/dL). Unexpectedly, decreased in serum zinc was found in both groups (3.01 ± 3.24 mMol/dL and 3.12 ± 3.71 mMol/dL). There was significant higher increment (P=0.045) in body weight among the fortified milk group (1.31 ± 0.69 kg) as compared to the non-fortified group (1.13 ± 0.69 kg). Consistently, there was significant increase (P=0.025) in the indicator of underweight (WAZ) among the fortified milk group (1.47 ± 0.50) as compared to the non-fortified group (1.33 ± 0.47). There was significant improvement (P=0.001) of cognitive performance, i.e. coding test-score among the fortified group (12.74 ± 11.76) as compared to the non-fortified group (8.31 ± 9.60), but there was no significant difference found in the improvement of physical capacity score in both groups (10323.77 ± 9253.83 versus 8435.94 ± 8824.55).

Conclusion Among underweight schoolchildren aged 7-9 y, supplementation of milk fortified with iron and zinc can provide better growth, and better speed processing of learning ability. ***(Med J Indones 2009; 18: 193-202)***

Key words: *Iron and zinc, underweight, growth, physical capacity, cognitive function, schoolchildren*

Undernourishment is widely known to be related to poor food intake that is closely caused by poverty. This condition leads to a high prevalence of underweight, stunting and wasting, especially among children up till adolescent. Indonesian school-aged children, especially those from poor families were at most in these conditions that resulting in poor physical capacity and learning ability, i.e. cognitive performance.

Poor food intake and low nutrition knowledge may cause inadequacy of nutrient intakes, both macro- and micronutrients. Schoolchildren need sufficient nutrients to be able to grow and developing well, physically, mentally and for their learning ability. Those who are undernourished also suffered from micronutrient deficiencies.[1] Besides macronutrients, i.e. energy and protein, micronutrients such as calcium, iron and zinc are widely known nutrients that affect child growth, i.e. increase in weight, height and body composition which can influence physical fitness performance, child development, including cognitive performance. [2]

To overcome nutrient inadequacies or undernourishment among school-aged children, several approaches could be implemented, i.e. nutrient supplementation, fortified and/or non-fortified food supplementation with or without nutrition education. There are several studies regarding the positive and negative effects of nutrient supplementation to child growth and development, but in our knowledge, limited studies was found on its effects toward nutrient fortified food supplementation.

Meat, milk and eggs are amongst related food closely affecting child growth, because they mostly good sources of protein, calcium, iron and zinc. Among these mentioned foods, milk is the most acceptable by children and it is easily digested. In addition, it is a fact that milk provides almost all nutrients needed and can be well digested by the body. It is also frequently drink by the children. However, for high price reason as well as other reasons, after the age of two, most Indonesian children discontinued consuming milk. And, this could answer the phenomena of the high prevalence of undernourishment found among schoolchildren especially those belonged to poor families.

Iron deficiency anemia is still one of the main public health problems in several countries in the world, including Indonesia.[3-6] Iron deficiency alone among children and adolescence with and/or without anemia is strongly related to learning ability and school achievement, besides of its closed relation to growth and loss of appetite. [6-9]

Zinc, it is involved in metabolic processes, and it was stated from several studies that zinc deficiency greatly affecting body growth, development, and immunity.[4] Thus, the combination of iron and zinc enriched in milk powder might have a strategic role in the prevention of child growth and development impairment. The addition of ferric-pyrophosphate could increase iron store while the addition of zinc could improve child mid-upper arm circumference and body weight. And, in general, this combination has no negative effect, biochemically.[4,8-10]

Unfortunately, iron and zinc are mostly found in meat and seafood which are classified as expensive foods. Poor socio-economic status is affecting to the poor quality of poor iron and zinc intake from daily foods, especially of those of hem-iron sources. Thus, will increasing the morbidity level among children. Several studies showed positive effects of milk fortified with iron and zinc on the prevalence of anemia and physical growth.[11]

In general, studies aimed to alleviate iron and zinc deficiencies were mostly done for women (pregnant women, adult or puberty) and infants, and it is rarely done to schoolchildren. In fact, schoolchildren need sufficient nutrients to be able to grow and developing well, physically, mentally and for their learning ability. There are school aged children who are undernourished and having micronutrient (iron and zinc) deficiencies as well.[2] This study conducted using two-parallel, randomized, double-blind, controlled intervention study to evaluate the effect of supplementing iron-zinc fortified milk to underweight schoolchildren aged 7-9 years (preadolescent period) on their physical growth and capacity, and learning ability. The hypothesis was that the underweight children would benefit more from the enriched milk intervention.

METHODS

Study population

The study was conducted from July 2007 to February 2008 in schoolchildren aged 7-9 y from public elementary schools of lower socioeconomic status in the eastern-district of Jakarta and in the Central Java of Surakarta. Pre-adolescent age- period was intended to study to have homogenous slow constant growth among the subjects before the growth spurt during adolescent. Children should met the inclusion criteria for age, weight-for-age Z-score of less than -2SD (standard deviation), did not have any congenital disorders, renal diseases, thyroid, diarrhea, and thalasemia, and parents released their permission by signing the informed consent. However, children who were severely anemic (hemoglobin value of less than 8 mg/dL), or who did not comply toward research procedure or participating to other studies were excluded.

All schoolchildren from 5 public schools in Jakarta and 10 public schools in Surakarta located in the poor urban areas were screened for eligibility. A total of 245 children were randomly assigned in the study

Sample size calculation was done using 80% power, 5% of type I error to enable detection of an effect size of 9.0 SD for speed processing indicator (coding test score) and expected significant difference in coding test score between groups was 4, resulted a required sample size of 107 subjects per-intervention group. Ethical clearance to conduct the study was released from the Committee on Medical Research Ethics, Faculty of Medicine University of Indonesia, Jakarta.

Study design

This study was designed as a clustered-randomized, two-parallel group, double-blind controlled community trial. Subjects (n=245) were group-randomly allocated by school to an iron-zinc fortified milk-group (n=121) or to a non-iron-zinc fortified milk-group (n=124). Random assignment was done by means to fitting the number of children per-school. Run-in period was done for a week before intervention began, i.e. immediately after baseline data for nutritional, biochemical, physical capacity and cognitive functions were collected. The intervention was continued for 6 months.

Intervention

Two sachets of milk or 54 g per-day containing additional of 6.56 mg of iron and 2.38 mg of zinc (iron-zinc fortified milk) as compared to standard milk with of 0.26 of iron and 0.88 mg of zinc were given to participants (Table 1).

The milk-supplements were indistinguishable in package and color and were coded as milk-A and milk-B. The codes remained unknown to both investigators and participants until the study was completed, all data had been entered, and initial analyses had been performed.

Tabel1. Composition of the intervention products

Nutrition value perserving	27 g Fortified milk (A)	27 g Unfortified milk (B)
Energy (kcal)	100.0	100.0
Fat (g)	3.7	3.7
Protein (g)	4.0	4.0
Carbohydrate (g)	12.7	12.7
Glucose (g)	6.4	6.4
Sodium (mg)	59.0	59.0
Vitamin A (Iu)	396.0	396.0
Vitamin D 3 (Iu)	51.0	51.0
Vitamin E (Iu)	5.0	5.0
Vitamin K (mcg)	9.1	9.1
Vitamin C (mg)	50.0	50.0
B1 (mg)	0.09	0.09
B2 (mg)	0.31	0.31
B3 (mg)	1.43	1.43
B6 (mg)	0.14	0.14
B9 (mcg)	44.0	44.0
B12 (mcg)	0.4	0.4
Potassium (mg)	194.0	194.0
Magnesium (mg)	14.0	14.0
Zinc (mg)	1.19	0.44
Fe (mg)	3.28	0.13
Ca (mg)	179.0	179.0
Phosphor (mg)	120.0	120.0

The milk was to be consumed twice-daily. The study participants were required to prepare and consume the milk once at school prepared by the appointed teachers, and once at home under parental supervision. Compliance monitoring was confirmed from records of sachets issued and returned, along with self-report calendars from the teacher. During school holidays and the fasting month (Ramadan), the milk was consumed at home, and the parents were instructed that the milk were to be consumed twice-daily and only for the subject.

Nutritional status indicators

Weight, height and mid-upper arm circumference were measured at baseline, and at 3 and 6 mo using standardized measurements. Nutritional status indicators of weight-for-age, height-for-age and weight-for-height Z-scores were analyzed using Epi-Infor 2000 soft-ware. Nutritional status level was classified using cut-off of -2SD.

Biochemical indicators

Non-fasting venous blood samples were obtained at school. Separation of red blood cell and of serum or plasma was conducted by centrifuging the blood immediately (3000 x g, 15 min at room temperature), after which the serum was divided into aliquots. The hemoglobin concentration of the blood samples was assessed by absorption spectrophotometry (Cell-Dyn 3700; Abbott, Abbott Park, IL). Serum ferritin was measured by a Microparticel Enzyme Immunoassay (Abbott AxSYM analyzer). Serum zinc was analyzed by measuring zinc from serum lithium heparinized tubes by use of flame atomic absorption spectrophotometry (Perkin Elmer 5100PC; Waltham, MA).

Physical capacity test

Physical capacity test was measured at baseline and at 3 and 6 mo. Harvard step test modified with wearing a backpack containing bags of finery sugar equal to 20% of individual child's body weight.[12] Step-test score was calculated using the following formula:[12]
Score = duration of tests (in seconds) x 100 + (heart rate minute-1 + heart rate minute-2 + heart rate minute-3

The step-test assessment was evaluated by trained medical doctors. The tests were done in special classroom and free from distraction, and the assessment session took approximately 5-10 min.

Cognitive function tests

Cognitive performance tests were measured at baseline and at 3 and 6 mo. The cognitive assessment battery included: 1) coding test (speed processing); 2) digit-forward (attention and short-term memory); 3) digit-backward (attention and short-term memory); and 4) visual search (attention and short-term memory).[13] The cognitive assessment battery was administered to subjects by a registered licensed psychologist. The tests were done in special classroom and free from distraction, and the assessment session took approximately 60-90 min.

Data management

All data collected were edited for its completeness and consistencies, and entered into spread-sheet database using SPSS (version 11.0) statistical software package (Chicago, USA) for furthered data cleaning. Anthropometric data was analyzed to provide weight, height and mid-upper arm increments, and Z-scores for weight-for-age, height-for-age and BMI-for age using Epi-Info 2000 to be furthered classified into underweight, stunted and wasted status using -2SD cut-off. Nutrients intake was analyzed using Nutrisurvey program 2005 and furthered classified into deficient in certain nutrient against to the Indonesian RDA for children aged 7-9 y. Cognitive function tests were reviewed individually and scored by the psychologist.

Statistical analysis

Nutritional status, biochemical, physical capacity test and cognitive test outcomes
The effect of milk supplementation with or without iron-zinc fortification on change in anthropometric measures and nutritional status indicators, biochemical indicators, physical capacity test and cognitive test outcomes were analyzed using analysis of mean differences.

Normality test was conducted for each of variables to select appropriate statistical tests (parametric or non-parametric tests). Continuous data was analyzed by milk intervention using independent-t test or Mann-Whitney; and categorical data was analyzed using chi-square test cross-tabulated by milk intervention. To draw conclusion on the results of repeated measures throughout the study period, appropriate multivariate analysis was performed. All statistical analyses tests were performed using statistical program for social sciences (SPSS) version 11.0.

RESULTS

In both study locations (Jakarta and Surakarta), 259 underweight children were randomly allocated to the 2 intervention groups, and 95% (n= 245) underweight children completed the study (Figure 1). The 14 underweight children (5.4%) who did not complete all measurements (dropped out) during the study either had moved (n = 7), absence from school without any notice for more than a week (n = 4), or could not participate further because of family issues (n = 3). There was higher number of dropouts between intervention groups but there was similarity in the reasons for dropping out. Furthermore, underweight children who dropped out during the study had similarity in baseline characteristics from underweight children who completed the study (data not shown).

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All underweight children who completed the study were included in the analysis regardless of milk-drinking compliance. Compliance to the milk regimen assessed by returned sachet count and confirmed by check-list provided by the appointed school-teachers. The compliance was 80.63% (12.26%) for iron-zinc fortified milk group and 79.13% (15.94%). There was no significant difference in milk compliance between intervention groups (P = 0.407).

Baseline demographic, anthropometric, biochemical, physical capacity and cognitive performance characteristics of the underweight children in each intervention group are shown in Table 2. Underweight children in the iron-zinc fortified milk group had higher proportion of boys and higher score than of physical capacity test score those in the non iron-zinc fortified milk group. There is no other baseline difference between intervention groups nor between study-location were observed.

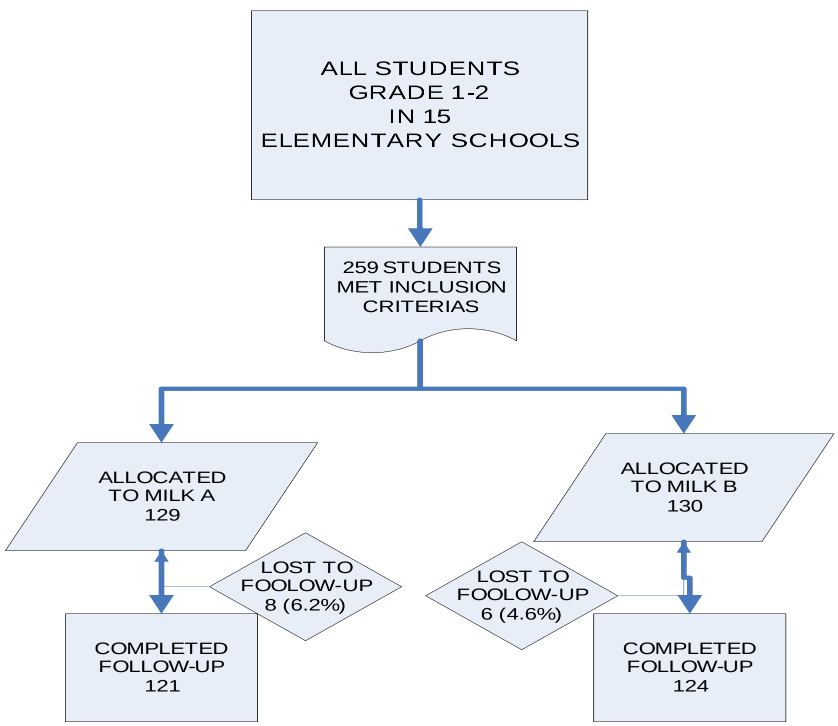

Figure 1. Flow chart of participants

Table 2. Baseline characteristics of subjects by assigned group, in mean + SD

	Zinc-iron fortified milk	Non zinc-iron fortified milk
N	121	124
Age (years)	7.94+0.57	7.95+0.55
Boys/Girls (no)	66/55	62/62
Anthropometric measures & indicators:		
Weight (kg)	18.02+1.48	18.10+1.42
Height (cm)	115.29+4.43	115.63+4.50
Mid-upper arm circumference (cm)*	16.34+0.89	16.57+0.79
Weight-for-age Z-score (WAZ)	-2.71+0.62	-2.66+0.63
Height-for age Z-score (HAZ)	-2.16+0.68	-2.10+0.73
Body mass index Z-score (BMIZ)	-1.8+50.75	-1.88+0.84
Underweight (n, %)	121 (100)	124 (100)
Stunted (n, %)	69 (57.0)	66 (53.2)
Wasted (n, %)	43 (35.5)	50 (40.3)
Biochemical indicators:		
Hemoglobin (g/dL)	13.00+0.95	12.88+0.97
Serum ferritin (mcg/dL)	30.97+22.38	34.22+23.85
Serum zinc (mMol/dL)	13.58+3.08	13.40+3.30
Anemia (n, %)	17+14.0	16+12.9
Iron deficiency (n, %)	14+11.6	16+12.9
Zinc deficiency (n, %)	14+11.6	15+12.1
Physical capacity test outcome:		
Harvard step test score	8963.88+4791.54	6794.27+3465.01
Cognitive test outcome:		
Coding score	27.35+11.13	27.33+11.06
Digit-span forward score	5.55+3.11	5.33+3.05
Digit-span backward score	2.59+1.47	2.65+2.03
Visual search score	3.85+7.18	2.28+13.69

Nutritional status indicators outcomes

Table 3 show that there were significant increases in weight, height and mid-arm circumference in both intervention groups. There were also significant improvements in nutritional status indicators in terms of weight-for-age, and height-for age Z-scores in both groups, but only BMI-for-age Z-score for the iron-zinc fortified milk group. The iron-zinc fortified milk improvements in weight and weight-for-age Z-score were significantly higher than were the non fortified milk.

Biochemical indicators outcomes

After 6 mo of intervention, there was positive effect of the biochemical iron status (hemoglobin and serum ferritin) but not for zinc status (serum zinc) in both intervention groups. There was no significant difference in the changes of biochemical indicators outcomes between the intervention groups.

Physical capacity outcomes

After 6 mo of intervention, there were significant positive effects of the physical capacity tests in both intervention groups. However, there was no significant difference in the increased of physical capacity score between the intervention groups.

Cognitive function outcomes

After 6 mo of intervention, there were significant increases in all cognitive function tests score in both groups. However, significant higher cognitive function test score improvement was only found for coding test reflecting for speed processing indicator among the iron-zinc fortified milk group as compared to the non iron-zinc fortified milk group.

Table 3. Effects of milk intervention on changes in nutritional status, biochemical and cognitive test indicators over 6 months in subjects by assigned groups, in mean (SD)

	Zinc-iron fortified milk	Non zinc-iron fortified milk	P-value
Anthropometric measures	n=121	n=124	
Weight change (kg/6 mo)	+1.31 (0.69)	+1.13 (0.69)	0.045
Height change (cm/6 mo)	+3.03 (1.00)	+2.85 (.88)	0.139
MUAC change (cm/6 mo)	+0.39 (0.62)	+0.24 (0.68)	0.080
HAZ change	+1.22 (0.42)	+1.18 (0.39)	0.467
WAZ change	+1.47 (0.50)	+1.33 (.47)	0.025
BMIZ change	+0.14 (0.46)	+0.07 (0.47)	0.200
Underweight change (n, %)	-22 (18.2)	-13 (10.5)	NA
Stunted change (n, %)	-9 (7.4)	-4 (3.2)	NA
Wasted change (n, %)	-5 (4.1)	0	NA
Biochemical indicators:			
Hemoglobin change (g/dL)	+0.01 (0.96)	+0.17 (0.81)	0.144
Serum ferritin change (mcg/dL)	+12.77 (25.50)	+14.99 (29.56)	
			0.531
Serum zinc change (mMol/dL)	-3.01 (3.24)	-3.12 (3.71)	0.806
Anemia change (n, %)	-7 (5.7)	-3 (2.4)	NA
Iron deficiency change (n, %)	-9 (7.5)	-9 (7.3)	NA
Zinc deficiency change (n, %)	+30 (-24.8)	+38 (-30.6)	NA
Physical capacity outcome:			
Harvard step test score change	+10323.77 (9253.83)	+8435.94 (8824.55)	0.103
Cognitive test outcome:			
Coding score change	+12.74 (11.76)	+8.31 (9.60)	0.001
Digit-span forward score change			
	+1.07 (3.15)	+0.88 (2.69)	0.602
Digit-span backward sore change	+1.10 (2.13)	+0.69 (2.30)	0.153
Visual search score change	+7.43 (7.01)	+7.67 (12.11)	0.849

Note : + : increase
- : decrease

DISCUSSIONS

Several trials have shown that combined iron and zinc supplementation has less of an effect on biochemical or functional outcomes than does supplementation with either mineral alone. However, there is no strong evidence to discourage combined supplementation, because supplementation programs that provide iron and zinc together are an efficient way to provide both micronutrients and provided the benefits of individual supplementation are not lost.[10] This study tried to evaluate the effectiveness of combined iron and zinc fortified in milk supplementation on biochemical, growth, and functional outcomes (physical capacity and cognitive performance) given daily to underweight school-aged children for six months.

The compliance of milk supplementation and micronutrient status

Compliance to drink the milk supplementation in both groups is similar, i.e. 80%. Iron status, i.e. anemia and iron deficiency among subjects in both groups at the beginning of the study were already low. After six months, there were no significant differences in the increased of hemoglobin and serum ferritin values among both groups. However, the 3.3% higher decreased in anemia prevalence after six months in the iron-zinc fortified milk group could be related to a positive effect of additional intake of iron in milk supplementation.

Iron deficiency is the main cause of anemia in all ages, and iron deficiency anemia is still major health-nutritional problem in Indonesia. It is mainly prevalent among pregnant women, under-five children and infants. However, it is also one of nutritional problem found among school-aged children, particularly among the poor.

Hemoglobin and serum ferritin are two of several diagnostic tools to determine iron deficiency besides of the varied clinical symptoms, and several laboratory findings, i.e. erythrocyte count and index, hematocrit, serum iron, total iron binding capacity, serum transferring, erythrocyte proto-porphyrin, and serum transferring receptor.[14,15]

In this study, it was unexpected to find that among the underweight subjects only approximately 13% had hemoglobin value of less 12 g/dL with approximately only 12% had serum ferritin value of less 12 mcg/dL. By observation, although there was poor environment sanitation and personal hygiene, however, the subjects were exposed to annual de-worming and iron pill program, and exposed to animal protein rich snacks during school break, such as meat balls or chicken nuggets. This finding was in accordance to Indonesian study finding among adolescent school girls that only found only 17% of anemic cases, and among the anemic subjects only 22% had iron deficiency anemia.[16]

Similar to iron status, biochemical data result shows that zinc deficiency was not prevalent among the underweight school-aged subjects. Unexpectedly, milk supplementation with or without iron-zinc fortification were both decreased serum zinc and consistently increased the prevalence of zinc deficiency. Furthermore, serum zinc level and zinc deficiency prevalence were found to be worse in the non iron-zinc fortified milk group. This finding is in accordance to the fact that there is zinc homeostasis within the body to regulate normal serum status. When dietary zinc intake is reduced or zinc needs are increased due to growth, the individual goes into negative zinc balance for a period of time before zinc balance is re-established at the lower level of intake. Increase intakes of several nutrients such as provided in the milk supplement, theoretically will decline serum zinc levels progressively in association with hormonal changes and tissue uptake of circulating nutrients induced by fuel metabolism. Thus, normal zinc status does not automatically exclude possibility of zinc deficiency. And it is suggested to use other marker for assessing risk of zinc deficiency in population, includes several indicators such as prevalence of stunting, adequacy of dietary zinc intake, biochemical measures of zinc concentration, and measurement of functional responses.[17]

We need to perform further laboratory analysis in order to be able to explain the the decreased in serum zinc but increased in serum ferritin found in this study, because the result clearly showed that the iron-zinc ratio in both milk do not influence the micronutrient interaction. A review of several studies of various ratios among adults that showed the degree of inhibition of iron on zinc uptake occurred by excess iron in the ratio of 2:1 or greater.[18] There were still unclear evidence on the improvement of iron and zinc status affected by iron-zinc supplementation with ratio 4:1.[19,20,21,22,23] or even with ratio 1:1.[4,24,25] An Indonesian study among adolescent school girls revealed that adding zinc to iron supplementation with ratio Fe:Zn = 2:1 or with ratio Fe:Zn = 4:1 protected the adverse effect of iron on decreasing zinc status 16. The study also stated that the competition between iron and zinc might occur in the role of both nutrients for erythrocytes formation as well as at the storage level, and supplementation with the ratio of Fe:Zn = 2:1 had minimal interaction on improving both iron and zinc status.

Effects of milk supplementation on growth

At the beginning of the study, both intervention groups had poor nutritional status. Approximately 55% and 37% of these underweight subjects also had stunting and wasting status, respectively. This finding shows a possibility of the more chronic or long-term onset of poor nutrition condition that usually blames poverty as the root of the problem resulting to protein-energy deficiency.

In this study, milk supplementation given to the underweight subjects in both groups significantly increased the calorie and protein intake by 200 kcal (≈10% of the recommended total calorie intake) and 8 g (≈17% of the recommended total protein intake), respectively. This might be a strong contribution to the improvement on growth indicators, i.e. increased of weight, height and arm circumference measures after six months. This finding could confirm the possibility that all subjects initially had limited calorie and protein intakes. However, this finding is not in accordance to the study done in Mexican preschoolers that found zinc and iron supplements had no effect on growth or body composition.[23]

Milk supplementation with or without iron and zinc fortification provides source of dietary protein, calcium and vitamin D that supporting child growth.[26] However, there are other essential minerals needed for child growth and development. Among other are iron and zinc which are mostly found in foods originated from animal products, such as meat and organs. Iron has function in oxygen transport as a component of hemoglobin in red blood cells and component of myoglobin (a muscle protein), and it is needed for certain reactions involving energy formation. On the other hand, zinc is required for the activation of many enzymes involved in the production of proteins, and as component of insulin and many enzymes. Thus, in relation to the initial condition of the subjects, zinc deficiency is closely related to growth failure, and loss of taste and appetite.

After six mo of intervention, higher increases in all growth indicators and higher reductions in the prevalence of underweight, stunting and wasting among subjects received iron-zinc fortified milk suggests that iron and zinc are probably mostly limiting nutrient among the study subjects. And, the interaction between iron and zinc in milk is an important factor in improving growth among underweight school-aged children. Positive growth response, in terms of increased weight and decreased underweight prevalence was significantly higher among underweight subjects receiving iron-zinc fortified milk supplementation that could be related to the higher intake of zinc. It is likely that young children would benefit from a combination of zinc supplementation and increased dietary intake which might lead to normalization of their anthropometric measurements more quickly. [28]

Effects of milk supplementation on physical capacity

At the beginning of the study, there was a significant difference in the physical capacity score between intervention groups. This might be due to the effect of clustered-randomization as compared to the individual-randomization. After six months, both of the intervention groups could increase the score significantly. However, the increased score was different significantly between groups. Higher score increase found in the iron-zinc fortified milk-group could explain that the study subjects not only had limitation in calorie and protein but also in micronutrient such as iron and zinc. Iron plays important role in energy metabolism and zinc plays important role in protein metabolism. Both, calorie and protein are important factors for physical capacity. In this study, the increased in mid-upper arm circumference measure found in both groups shows that there was increased in muscle mass and strength that was represented by improved physical capacity. This finding is supported by Sen and Kanani (2006) that concluded anemia is likely to adversely affect physical work capacity in young adolescent girls.[29]

Effects of milk supplementation on cognitive performance

After six mo of milk supplementation, there was a significant difference in speed processing test score increase between groups (i.e. coding test). In addition, beside visual-search test, other short memory and attention score tests conducted in this study (digit-span forward and backward tests) shows trend of higher increment among those receiving iron-zinc fortified milk supplementation. These findings show positive benefit of the interaction between iron and zinc in milk in improving cognitive performance among underweight school-aged children. Study of found that anemia is likely to adversely affect cognition in young adolescent girls. [28]

Serum zinc was significantly lower in malnourished children as compared to well-nourished children.[29] Zinc, is essential nutrient that will be supplied to the brain via both the blood-brain and blood-cerebrospinal fluid barriers. It is most concentrated in the limbic system, i.e. the hippocampus and amygdale, as zinc-containing glutaminergic neuron-rich areas. Dietary zinc deprivation may influence zinc homeostasis in the brain, resulting in brain dysfunction such as learning impairment.[30]

It is concluded that supplementation with or without iron-zinc fortification consumed twice daily for six months improved nutritional, biochemical, and functional status (physical capacity and cognitive function) in underweight school children aged 7-9 y. In addition, although there was no significant biochemical status difference between the intervention groups, iron and zinc fortified in milk had superiorities in significantly improving weight, weight-for-age Z-score and coding test score (speed processing performance) in underweight school children aged 7-9 y.

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