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Assessment of Sodium Content of Processed Food Available in Indonesia

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ABSTRACT

This study was conducted to identify the sodium content in processed food and determine the proportion of products that meet the World Health Organization (WHO) global sodium benchmark criteria. Spesific concern was placed for sodium content in instant noodles. A comparative analysis was conducted to determine the relevance between the WHO and Indonesian Food and Drug Authority (Indonesian FDA) benchmarks in limiting sodium in instant noodles. Data on sodium levels (mg/100 g) of processed food was obtained from the register in the Indonesian FDA, Directorate of Processed Food Registration for 2019 to 2020. There were 3,850 products, consisting of 3,036 Local Products (LP) and 814 Imported Products (IP). These products were grouped into seven food categories and 18 types of food. The highest sodium content was found in chili sauce at 2,254,06 mg/100 g, and the lowest was in wafers at 218.64 mg/100 g. Overall, 2,538 of all products (66.56%) did not meet the sodium criteria based on the WHO benchmark. While for instant noodles, only 14.2% of the products met the sodium criteria based on the Indonesian FDA regulations and only 7.4% comply to the WHO sodium benchmark. The Cohen's Kappa test showed a strong agreement (K=0.650; 95% CI; p=0.00, strong) between the two regulations in limiting sodium levels in instant noodles. This study provides an overview of sodium levels in processed food in Indonesia. The sodium content in most products including instant noodles, as one of the most frequently consumed products, are still above the recommended value. Therefore, it is necessary to develop sodium benchmark of wider range of food categories in national level that might contribute to sodium intake, as well as for instant noodles. In order to achieve this goal, involvement of multi stakeholder among government, food industry and expert are also needed to deliver effective policies regarding sodium intake concerns.

Keywords: assessment, benchmarks, processed food, sodium, WHO

INTRODUCTION

Salt plays an important role in processed food formulation by enhancing aroma and reducing the bitterness of a food product. In human body, sodium in salt is needed to transmit nerve impulses, muscle relaxation, regulates osmotic pressure and maintains water balance (De & De 2019). Sodium in food can be naturally contained or added to food through household cooking, processed and eating out food (Zhang *et al.* 2015).

The results of the 2014 Individual Food Consumption Survey (IFCS) analysis showed that the average salt intake of the Indonesian population reached 6.68±5.85 g/day exceeding the government recommendation of 5 g/day (Atmarita *et al.* 2017). Excessive sodium intake can be associated with increased prevalence of Non-communicable Diseases (NCDs) such as hypertension and cardiovascular diseases (Grillo *et al.* 2019). Meanwhile, the Indonesian 2018 Basic Health Research (BHR) showed that the number of people with hypertension had increased from 25.8 to 34.1%. This result can also be associated with high sodium intake (MoH RI 2013; MoH RI 2018a).

The correlation between NCDs prevalence and excessive salt intake urges the Government of Indonesia to issue a 2013 Minister of Health regulation concerning the inclusion of sugar, salt, and fat information and health messages in processed and prepared foods that recommends sodium intake of <2 g/day which is equivalent to salt <5 g/day. As WHO direction, its member countries are suggested to strengthen food and nutrition policies, one of which is mandatory nutrition labeling on processed foods (WHO 2018).

In Indonesia, the WHO directive is carried out through the issuance of Indonesian FDA

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No. 22 of 2019 concerning Nutritional Value Information which regulates the obligatory inclusion of nutrition facts in processed food label. Although the research conducted by Mauludyani *et al.* (2021) showed that 69.4% of respondents rarely read the information on the label, the inclusion of nutrition facts is necessary to increase consumer awareness about the nutrient content of the food they consumed.

In addition, the Indonesian FDA regulation No. 22 of 2019 also regulates nutrient profiles as a requirement for voluntary inclusion of the "healthier choice" logo on the labels. As an initiation stage, the nutrient profile only sets a limit on the sodium content of instant noodles, which is ≤900 mg/100 g (Indonesian FDA 2019). This decision on the limitation of sodium content in instant noodles was based the 2014 IFCS analysis which showed that instant noodles is a processed food that provides the largest contribution to the sodium intake of the Indonesian population in all age groups by 11.78% (MoH RI 2018b).

Due to the large contribution of processed food in sodium intake, it is important to identify sodium levels in various processed food (Nieto *et al.* 2018). This data is expected to help identify interventions needed to reduce sodium intake from processed foods. It also provides an opportunity for manufacturers to reformulate their products by reducing the sodium added to their products (Kamis *et al.* 2015).

Therefore, this study was conducted to analyse the proportion of food products that comply with WHO sodium benchmark targets (WHO 2021). An assessment of the sodium content of instant noodles was also carried out refering to sodium criteria set by the government and WHO. In addition, comparative analysis was conducted to determine the relevance and classification of the two benchmarks in limiting sodium contents in instant noodles. The data from this study might be beneficial for public health officials in developing strategies to reduce and monitor sodium trends in the food supply chain.

METHODS

Design, location, and time

This is a cross sectional descriptive study, utilizing a secondary data on sodium content of processed food products registered in the Indonesian FDA. Data collection and analysis were carried out in Jakarta from March to May 2021.

Sampling

Processed food in this study refers to food or beverage that is processed in a certain way or method with or without additional ingredients. The sample for the study was data from processed foods that can contribute to sodium intake. Data were obtained from processed food products registered in the Indonesian FDA trough the Directorate of Processed Food Registration, from 2019 to 2020. The variables included on this assesment were food category, type of food, sodium content as well as product origin. The product origin divided into Local Products (LP) and Imported Products (IP). There were 3,850 products' data used in this study, consisting of 3,036 LP and 814 IP products.

Data collection

Data on sodium levels were obtained from the database of the nutritional value of processed foods registered through the e-registration (e-reg) of the Indonesian FDA. Data was collected based on the distribution approval number, registration number, the type of food, and its sodium content. Data with the same marketing approval number were excluded to avoid duplication. The processed food data collected was grouped into seven food categories and specified into 18 types of food (described in the brackets) based on Indonesian FDA Regulation No. 34 of 2019 on Food Categories.

The Food category are as follow: food category 01.0 (dairy products and their analogues (processed cheese and cheese)), food category 06.0 (cereals and cereal products (cereals and instant noodles)), food category 07.0 (bakery products (cookies, wafers, crackers, white bread, and sweetbreads)), food category 08.0 (meat and meat products including poultry and game meats (processed meat and chicken)), food category 09.0 (fish and fishery products including molluscs, crustaceans, and echinoderms (processed fish)), food category 12.0 (salt, spices, soups, sauces, salads, and protein products (soy sauce and chili sauce)), and food category 15.0 (ready-to-eat snacks (savoury snacks, chips, extruded snacks, also nuts and their products)). Then, the processed foods studied would be described as 18 types of food. The data presented is the sodium content of "as sold" food product in mg/100 g units.

Data analysis

Data analysis was carried out descriptively using SPSS 22 software to identify the median, interquartile range, minimum and maximum values of sodium content for each type of food. Statistical analysis using the Mann-Whitney test ($p<0.05$) was conducted to determine the difference in sodium levels among LP and IP products. The proportion of products that exceeded the WHO sodium benchmark and sodium criteria for instant noodles based on the Indonesian FDA Regulation No. 22 of 2019 was set out and presented descriptively.

Cohen's Kappa ($p<0.05$) test was conducted to determine the agreement between the Indonesian FDA regulation and WHO global sodium benchmark in classifying the sodium content in instant noodles. Kappa score is a statistical ratio of categorical data that describes the proportion (%) of the number of measurements that are consistent (similar) between two raters compared to the proportion (%) of the number of measurements that differ between two raters. The kappa value result ranges from -1.0 to 1.0 and the level of agreement can be determined as follow ≤0.20 (poor), ≤0.21−0.40 (fair), ≤0.41−0.60 (sufficient), ≤0.61−0.80 (strong), and ≥0.81 (very strong). The greater the kappa coefficient, the stronger the relevance between the two raters in classifying the recommended dietary sodium criteria.

RESULTS AND DISCUSSION

Processed food that potentially become a contributor to sodium intake

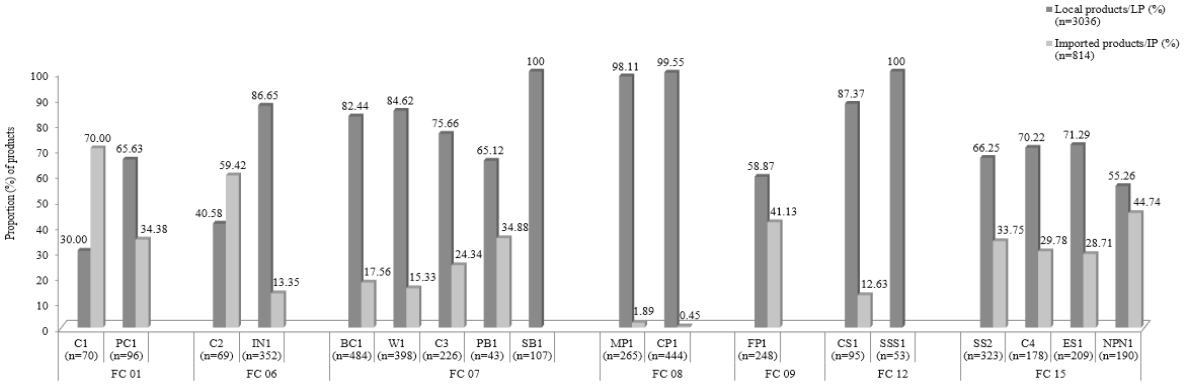
There were 3,850 processed food products identified as contributor to sodium intake collected from the e-reg database. The data was divided into 3,036 (78.86%) local products (LP) and 814 (21.14%) Imported Products (IP). The results of food grouping were divided into seven Food Categories (FC) and 18 types of food. The proportion of products in each food category analyzed in this study is described in Figure 1.

For sweet bread and sweet soy sauce, the data is only represented by local products because there is no imported product in the database for those types of food. However, for cheese and cereals, the proportion of Imported Products (IP) is higher than local products by 70 and 59.42%, respectively. Overall, the largest number of product samples was from meat and meat product category (FC 08) with as many as 709 products. Meanwhile, white bread had the smallest number with only 43 products. The types of foods identified were almost the same as the assessment of sodium levels in processed food conducted in Malaysia (Haron *et al.* 2020) which included processed meats (fish, chicken, and beef), breakfast cereals, sauces, cheeses, savory snacks, sweet snacks, and white bread.

Sodium levels in 18 types of processed foods registered in the Indonesian FDA from 2019 to 2020

The top three in the highest average of sodium content regardless of the product origin is shown in chili sauce (1,854 mg/100 g), soy sauce (1,319 mg/100 g), and instant noodles (1,275,31 mg/100 g). Similar results was also found in Brunei Darussalam by Kamis *et al.* (2015) where the sauces and seasonings showed the highest sodium content among other types of food. Analysis conducted by Prihatini *et al.* (2016) showed that sodium intake in the age group 6−18 year from sauces products were 7.9%, which was ranked second after instant noodles (13.2%). Study by Amarra and Khor (2015) from the 2012 National Socio-Economic Survey (SUSENAS) stated that sodium intake per capita in Indonesia from the consumption of instant noodles, sweet soy sauce and chili sauce were 144.13, 115.32 and 3.43 mg/day, respectively. In line with this, based on the 2014 Individual Food Consumption Survey (IFCS) analysis in DKI Jakarta province conducted by Setyowati *et al.* (2018) showed that the average daily consumption of those three types of food was 36.25, 4.02 and 1.76 g/day in all age groups of the population.

There was a significant difference ($p<0.05$) between sodium levels in LP and IP products found in processed cheese, instant noodles, biscuit and cookies, wafers, crackers, white bread, chips, and processed nuts. Sodium content tends to be higher in local products rather than imported products for processed cheese, biscuits, wafers, crackers and nuts. Whereas in instant noodles, white bread and chips, the sodium content were tended to be lower in local products compared to imported products. The average sodium content in each food types and the product origin are depicted in Table 1.



Note: Food category 1 /FC01 (C1: Cheese; PC1: Processed cheese); FC06 (C2: Cereals; IN1: Instant noodle); FC07 (BC1: Biscuit/Cookies; W1: Wafers; C3: Crackers; PB1: Plain bread; SB1: Sweet bread); FC08 (MP1: Meat processed; CP1: Chicken processed); FC09 (FP1 : Fish processed); FC12 (CS1 : Chili sauce; SSS1 : Sweetsoy sauce); FC15 (SS2: Savoury snacks; C4: Chips; ES1: Extrudate snacks; NPN1: Nut and processed nuts)

Figure 1. Proportion (%) type of food products contributing to sodium intake in each food category (FC)

The proportion of products meeting the WHO global sodium benchmark

In general, the median sodium levels of the food products exceeded the limits set by WHO, except for wafers, crackers, and cereals (Table 2). Their proportion that met the WHO sodium criteria were 73.87, 68.14, 56.52%, respectively. These results were followed by sweet breads (48.59%), extruded snacks (45.93%), cheese (45.71%), and nuts and their processed products (45.26%). WHO stated that if more than 45% of products have met the target, then the sodium level threshold tends to be loose and can be lowered to become more stringent (WHO 2018).

Six products with the least proportion of meeting the criteria of the WHO global sodium benchmark were shown in processed fish (2.42%), chili sauce (6.32%), instant noodles (7.39%), white bread (9.30%), processed cheese (14.58%), and savoury snacks (15.17%). According to Rosewarne *et al.* (2020), if the number of products on the market has met the criteria of around 33%, with a range of 10% above or below (23−43%), the sodium content limit tends to be feasible and appropriate. In the other hand, WHO stated that if less than 10% of existing products meet the criteria, then the sodium level threshold can be increased/loosened. So that, it is easier and more feasible to achieve (WHO 2018). It is important to set the sodium threshold that is appropriate to sodium content of products that are available in the national market. Thus, setting national benchmarks is more recommended rather than an approach to reach international benchmark. Still, the percentage of products that complied with WHO sodium benchmark could be considered as an overview of reference point regarding feasibility and later as comparison of sodium criteria across food category in national level (Kloss *et al.* 2015).

The proportion of products that meet the WHO global sodium benchmarks criteria can be seen in Table 2.

The difference between the WHO global sodium benchmark and the sodium content of processed foods

The sodium content used as a reference is the median value gained from the descriptive analysis. It better represented the distribution of abnormal data obtained in this study (Rahman 2020).

The percentages (%) of sodium levels that are the most different from the WHO benchmarks are shown in chili sauce and processed fish at 187.54 and 176.57%, respectively. Those numbers exceeded almost twice the sodium limit recommended by WHO. Analysis of sodium levels in chili sauce in Malaysia conducted by Shahar *et al.* (2019) showed that the average sodium content in chili sauce was 1,240 mg/100 g. Judging from its conformity with the WHO sodium benchmark, there is a difference of about 90.77%, which is still smaller than what was found in this study. This is different from the analysis of sodium levels of chili sauce in Brunei (Kamis *et al.* 2015), which showed the average sodium content in chili sauce was 914

Table 1. Sodium content of various types of processed food registered in Indonesian Food and Drug Authority from 2019 to 2020

Type of food	n Total	Total Median (IQR) Min−Max	n (LP)	Local products Median (IQR) Min−Max	n (IP)	Imported products Median (IQR) Min−Max
Cheese**	70	689.23 (358.81)	21	707.84 (160.05)	49	686.32 (513.18)
		49.23−2,167,60		140.94−1,534,09		49.23−2,167,60
Processed cheese*	96	1,207,60 (716.13)	63	1,390,14 (570.81)	33	809.73 (505.12)
		86.31−3,225,50		86.31−3,225,50		149.64−1,600
Cereal**	69	230 (208.63)	28	203.39 (148.52)	41	296.00 (243.64)
		10.38−1,093,16		93.90−604,203,39		10.38−1,093,16
Instant noodle*	352	1,275,31 (545.33)	305	1,258,22 (475.24)	47	1,692,28 (1,112,88)*
		309.98−3,645,37		405.67−3,359,50		309.98−3,645,37
Biscuit cookies*	484	307.08 (154.12)	399	308.42 (138.87)	85	276.02 (213.19)
		16.68−1,197,02		16.68−1,197,02		44.45−600.00
Wafers*	398	169.66 (155.51)	337	180.47 (145.06)	61	105.59 (113.50)
		2.93−1,144,39		51.61−1,144,39		2.93−870.30
Crackers*	226	507.97(223.63)	171	525.00 (267.85)	55	453.77 (180.55)
		15.44−1,569,50		15.44−1,301,51		195.71−1,569,50
Plain bread*	43	446.22 (137.10)	28	422.84 (79.39)	15	617.58 (268.14)
		235.71−1,035,50		235.71−559.93		384.87−1,035,50
Sweet bread	107	313.84 (154.08)	107	313.84 (154.08)	0	-
		84.80−737.42		84.80−737.42		
Processed meat**	265	711.58 (405.78)	260	714.79 (404.64)	5	572.61 (568.32)
		40.72−4,680		40.72−4,680		439.46−1,181,12
ProcessedChicken**	444	621.46 (320.23)	422	621.46 (321.82)	2	616.14 (-)
		25.31−2,276,90		25.31−2,276,90		484.00−748.28
Processed fish**	248	746.73 (320.01)	126	750.02 (400.97)	102	745.95 (274.36)
		50.63−4,565,20		50.63−1,749,98		314.15−4,565,20
Chili, tomato sauce**	95	1,868,98	82	1,868,98 (986.90)	12	1,980,03 (3,912,47)
		(1,041,57)		421.26−8,456,86		263.73−5,374,10
Sweet soy sauce	53	1,319,00 (713.77)	53	1,319,00 (713.77)	0	-
		124.55−3,288,06		124.55−3,288,06		
Savoury snacks**	323	736.83 (584.76)	214	760.32 (566.90)	109	711.39 (619.75)
		33.18−2,131,36		33.18−1,799,00		97.74−2,131,36
Chips*	178	387.44 (340.24)	125	364.13 (315.63)	53	560.00 (384.09)
		15.82−1,490		15.82−1,253,89		89.98−1,490,00
Extrudate snacks**	209	564.60 (606.08)	149	627.81 (657.09)	60	481.03 (455.90)
		14.17−1,845,72		25.00−1,845,72		14.17−1,750,00
Nut and processed nuts*	190	303.69 (418.15)	105	346.36 (395.77)	85	215.00 (399.28)
		0.00−1,505,41		2.81−1,505,41		0.00−946.81
Total	3,850		3,036		814	

*p<0.05; ** p>0.05; LP: Local products; IP: Imported products

Table 2. Proportion of products that meet sodium limit of WHO Global Sodium Benchmarks

Type of food	n	WHO global sodium benchmarks (mg/100 g)	Median of sodium content (mg/100 g)	(n) comply	% comply
Cheese	70	≤625	689.23	32	45.71
Processed cheese	96	≤720	1,207,60	14	14.58
Cereal	69	≤280	230	39	56.52
Instant noodle	352	≤770	1,275,31	26	7.39
Biscuit, cookies	484	≤265	307.08	147	30.37
Wafers	398	≤265	169.66	294	73.87
Crackers	226	≤600	507.97	154	68.14
Plain bread	43	≤320	446.22	4	9.30
Sweet bread	107	≤310	313.84	52	48.59
Processed meat	265	≤540	711.58	227	32.02
Processed chicken	444	≤600	621.46	205	46.17
Processed fish	248	≤270	746.73	6	2.42
Chili, tomato sauce	95	≤650	1,868,98	6	6.32
Sweet soy sauce	53	NA	1,334,06	NA	NA
Savoury snacks	323	≤500	736.83	76	15.17
Chips	178	≤500	387.44	114	64.04
Extrudate snacks	209	≤520	564.60	96	45.93
Nut and processed nuts	190	≤280	303.69	86	45.26
Total					33.44

WHO: World Health Organization

mg/100 g, resulting in a difference of about 40.62% compared to the WHO benchmark. This indicates that the sodium content of chili sauce in Indonesia tends to be higher than other Southeast Asian countries, such as Malaysia and Brunei Darussalam.

The large difference in sodium levels in processed fish products was due to the WHO benchmark being low for that product category, which is 270 mg/100 g. This limit is very strict compared to the limits set by the Healthier Choice Logo (HCL) in Singapore and the WHO Regional Office for Europe nutrition profile of 550 and 680 mg/100 g, respectively (HPB 2020; Bonsmann *et al.* 2019). Thus, it needs to be discussed further considering that fishery products can contain sodium naturally. Therefore, a more comprehensive consideration is needed in setting the boundaries.

Based on Figure 2, a large difference was also seen in processed cheese (67.72%) and instant noodles (65.62%). The sodium limit set by the WHO benchmark for processed cheese was 720 mg/100 g, which was much lower than the median sodium content of locally processed cheese, which was found to be 1,390,14 mg/100 g. Imported processed chese complied better with this limit, with the average sodium content of 809.73 mg/100 g (see Table 1). Similarly, the sodium content of instant noodles (1,275,31 mg/100 g) was also higher than the sodium limit recommended by the WHO benchmark of 770

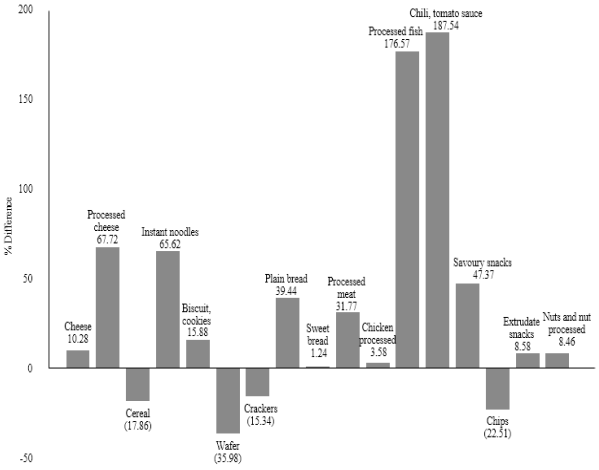


Figure 2. Difference (%) between sodium limit on WHO global sodium benchmarks and median value of sodium content

mg/100g. Compared to an assessment of instant noodles conducted in Malaysia, the median sodium content in that study was 1,800 mg/100 g (4.5 g/100 g salt) (Tan *et al.* 2019), resulting in a greater % difference of 133.77% sodium limits from the WHO benchmarks. This indicates that greater efforts are needed to reduce sodium levels in instant noodles. Despite the larger percentage difference to the WHO limit for processed cheese as compared to instant noodles, the average consumption of cheese (0.6 g/day) is still much lower than the consumption of instant noodles (36.25 g/day) based on IFCS data in DKI Jakarta (Setyowaty *et al.* 2018). Thus, limiting the sodium content of cheese may not be a priority given that its consumption tended to be low.

Mean and range of sodium content of instant noodles based on preparation method

In this assessment, instant noodles are grouped based on different preparation methods. The grouping also distinguishes the product's origin between LP and IP (Table 3). Based on the Mann-Whitney analysis ($p<0.05$), there was a significant difference in sodium content between the LP and IP instant noodles. As it is shown in Figure 3, the sodium content of the IP instant noodles soup tended to be higher than that of the LP product. While for fried instant noodles was lower than for IP product with 688.27 and 1,093,12 mg/100 g, respectively. The median sodium content of instant noodles was higher (1,406,43 mg/100 g) than fried instant noodles (1,083,05 mg/100 g). The same results were shown in an assessment conducted by Tan *et al.* (2019) for instant noodles in Malaysia, with average sodium levels in instant noodles soup and fried instant noodles were 1,828 and 1,136 mg/100 g, respectively, which were slightly higher than this study.

The proportion of instant noodles products that met the criteria for sodium requirements based on Indonesian FDA Regulation No. 22 of 2019

There were 14.20% of products that met the sodium criteria for instant noodles based on the Indonesian FDA Regulation No. 22 of 2019 (Table 4). For instant noodles soup, the proportion of products that met the criteria was 8.33%, smaller than fried instant noodles with 25%. There has been no similar research before. However, the small proportion of products (<10%) that met the sodium criteria for instant noodles soup was an indicator for instant noodles manufacturers to put greater efforts to meet these requirements (WHO 2018).

Data on sodium content can be used as the basis for determining nutrient profile thresholds that can stimulate reformulation of sodium levels by food manufacturers (Ahuja *et al.* 2019; Nieto *et al.* 2018). In determining the nutrient profile, it is very important to consider a realistic and achievable threshold for research and development that are relevant to nutritionists recommendations. Limits that are too tight make it difficult to reformulate (Lehmann *et al.* 2017).

Efforts on sodium content reduction had also been carried out in Kuwait. This had been conducted as a respond to lowering sodium intake through bread consumption, which is the main food contributor to sodium intake in the country. Therefore, the bread supplier in the market were encouraged to decrease its sodium content gradually every six months by 20% in 2013. Other mediterranean countries, such as

Table 3. Sodium content of instant noodles registered in Indonesian Food and Drug Authority from 2019 to 2020

Type of instant noodle	n (Total)	Total Median (IQR) Min−Max	n (LP)	Local products Median (IQR) Min−Max	n (IP)	Imported products Median (IQR) Min−Max
Instant noodles (soup & fried)*	352	1,275,31 (545.33)	305	1,258,22 (475.24)	47	1,692,28 (1,112,88)
		309.98−3,645,37		405.67−3,359,50		309.98−3,645,37
Instant noodles (soup)*	228	1,406,43 (629.42)	185	1,370.68 (534.76)	43	1,784,86 (1,005,85)
		309.98−3,654,37		493.01−3,359,50		309.98−3,645,37
Instant noodles (fried)*	124	1,083,05 (344.43)	120	1,093,12 (337.83)	4	688.27 (311.90)
		405.67−1,618,67		405.67−1,618,67		623.95−1,039,81

*$p<0.05$; LP: Local products; IP: Imported products

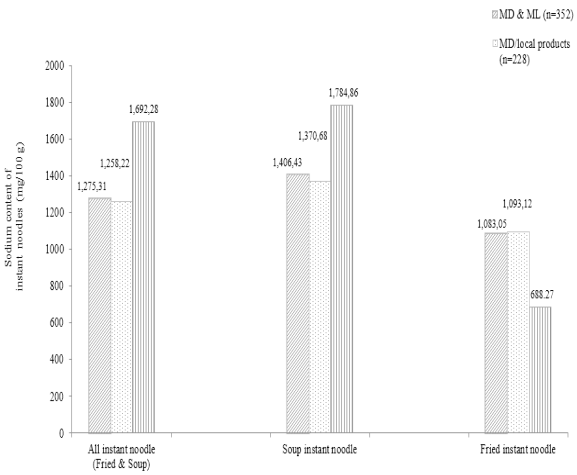


Figure 3. Median sodium content of instant noodles based on method of preparation

Table 4. Proportion of instant noodle products that met the sodium profile criteria based on Indonesian Food and Drug Authority regulation No. 22 of 2019

Type of instant noodles	n	Sodium nutrient profile by IFDA regulation (mg/100 g)	(n) comply	% comply
Instant noodles (soup & fried)	352	≤900	50	14.20
Instant noodles (soup)	228	≤900	19	8.33
Instant noodles (fried)	124	≤900	31	25

Jordan and Oman, have also prepared legislation by developing sodium benchmarks that become a reference for food products that are widely consumed in the region (Al Jawaldeh *et al.* 2018).

Comparison of sodium threshold by Indonesian FDA regulation and WHO global sodium benchmarks for instant noodles

Comparative analysis of sodium requirements in these two references revealed that the proportion of products that met the criteria based on the nutrient profile of government regulation was greater (14.20%) compare to the WHO global sodium benchmark (7.4%) (Table 5). However, the two benchmarks have strong consistency and agreement based on the Kappa Cohen's analysis. This indicates almost the same classification of the two regulations in limiting sodium levels in instant noodles. There have been no published studies that specifically compare the two benchmark models for instant noodles products. However, the same method is commonly used to compare two different nutrient profile models in order to determine the suitability or reliability of the nutrient profile model used with other nutrient profile models as a reference. This was demonstrated in other studies comparing models of varied nutrient profile with different types of food (Pivk Kupirovič *et al.* 2020; Labonté *et al.* 2017). This comparison can help regulators to determine the nutrient profile model to be used as a reference (Poon *et al.* 2018).

Eventhough the two benchmarks have strong agreement based on the Kappa Cohen's analysis, the national sodium criteria for instant noodles is more attainable. It is reflected by the fact that more products are able to comply with the national regulation compared to WHO sodium benchmarks. Since the national regulation on limiting the sodium content of instant noodles is implemented voluntarily, there would be slow progress towards lowering the Indonesian sodium intake at population level. Thus, more involvement of food industry are necessary to reach the goal. In the other hand, increasing consumer awareness of health risk of high sodium intake also remains important (Kloss *et al.* 2015).

Table 5. Comparison between the sodium criteria of instant noodle based on WHO gobal sodium benchmarks and Indonesian food and drug authority regulation No. 22 of 2019

Type of instant noodles	n	% comply by IFDA regulation	% comply by WHO benchmark	Value of kappa	Level of agreement	p
Instant noodles (soup & fried)	352	14.20	7.4	0.650	strong	0.00
Instant noodles (soup)	228	8.33	5.3	0.759	strong	0.00
Instant noodles (fried)	124	25	11.3	0.553	sufficient	0.00

IFDA: Indonesian Food and Drug Authority; WHO: World Health Organization

CONCLUSION

Data on sodium content can be an important basis for identifying sodium levels in processed foods available in the Indonesian market. The study found that most food categories had high sodium level, exceeding the WHO global sodium benchmark. Thus, it is necessary to set attainable sodium threshold recommendation that could be applied at national level. Comparative analysis between the two (national and WHO) sodium benchmarks set for instant noodles showed strong relevance in limiting the sodium content of instant noodles. Nevertheless, only few that meet the sodium benchmark set by both regulation. Currently, the initiative to stimulate a voluntary reformulation of sodium levels, only applied for instant noodles through the "healthier choice" logo on labels. Therefore, it is recommended to develop sodium benchmark for broader range of food categories that are identified as main contributor to sodium intake in the population. Through this mechanism, a multi-stakeholder coordination between the government, nutrition experts, and food producers is pivotal in formulating effective public policies and strategies regarding this objective.

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DECLARATION OF INTERESTS

The authors declare that there is no conflict of interest with other person or institution.

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Nutrition Fact Panel Use and its Association to Diet Quality among University Students in Universitas Indonesia

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ABSTRACT

The aim of this study was to assess the association between Nutrition Fact Panel (NFP) use and diet quality among university students. A comparative cross sectional study was conducted in 2019 among 172 college students living in dormitory of Universitas Indonesia, Depok, Indonesia. Data about socio-economic and demographic characteristics, food preparation, NFP use and nutritional knowledge were collected through interview using structured questionnaire. Nutritional status was obtained by direct height and weight measurement. Diet quality was measured using Diet Quality Index-International (DQI-I) score, derived from 3x24 hours recall. Finding of this study showed that the characteristics of NFP users and non-users were not differed significantly except for nutritional knowledge in which NFP users were 1.852 times more likely to have good knowledge than NFP non-users ($p<0.05$; 95% CI:1.009−3.396). The median total diet quality score was categorized as low (44 out of 100). There was no association between NFP use and total diet quality score even after controlling for possible confounder. However, the exploration in each component of diet quality measurement showed adequacy score of calcium was significantly higher in NFP users than non-users ($p<0.05$). NFP use also positively associated to total variety score ($\beta=0.985$; $p<0.05$). Future nutrition interventions could focus on enhancing the use and understanding of NFP among university students.

Keywords: diet quality index-international, nutrition fact panel, nutritional knowledge

INTRODUCTION

University students have been reported to have poor diet quality across countries. A systematic review of 37 studies conducted in Europe, North America, Middle East, South America and Africa found that most university students were likely to consume more fast food, sugar sweetened beverages, sweet and salty packaged snack but less vegetable and fruit (Bernardo *et al.* 2017). Similar situation was also found among university students in Indonesia. A study in Jakarta found that 75% of university students consumed instant food, most of which were instant noodle (Surjadi 2013).

Poor diet quality potentially lead to nutrition related health problem such as obesity, coronary heart disease, and type 2 diabetes mellitus (World Health Organization (WHO) 2018). Therefore, a preventable action need to be taken to improve dietary habit among university students whom are the potential group to adopt healthier dietary habits which might have a lasting impact in later life (Cooke & Papadaki 2014).

Considering the high exposure of university students toward packaged food, Nutrition Fact Panel (NFP) use could be considered as a simple yet rational preventive effort for improving dietary habit since it is available on most packaged food. NFP, which provide information needed by consumer for identifying healthier food option through the comparison of the food's nutrient content, might become an assisting tool at the time of purchasing (Viola *et al.* 2016). In Indonesia, the importance to read NFP was one out of ten messages in Indonesian balance nutrition guideline (Ministry of Health Republic of Indonesia (MoH RI) 2014).

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Finding from a cross sectional study among university students in Turkey showed that NFP users had higher dietary quality score compared to non-users (Buyuktuncer *et al.* 2018). Consistently, another study of college students in Minnesota, United States, had found NFP users associated significantly to healthier dietary practices such as higher consumption of fruit, vegetable and fiber but lower intake of fat and added sugar (Graham & Laska 2012).

However, studies about NFP use and its association to diet quality are largely conducted in developed countries where consumption of packaged foods were relatively high (Buyuktuncer *et al.* 2018; Graham & Laska 2012). To authors' knowledge there is currently limited empirical evidence related to this topic in low and middle income countries with different level of exposure to packaged food and different food habits. This study will use the context of Indonesia, a country categorized as low-middle income (World Bank 2021). Moreover, previous studies in Indonesia about food or nutrition label were mainly discussing the characteristics of label users (Mauludyani *et al.* 2021; Huda & Andreas 2016). Therefore, this present study aimed to investigate the association between NFP use and diet quality among university students in Universitas Indonesia.

METHODS

Design, location, and time

The current study was designed as comparative cross-sectional study in dormitory of Universitas Indonesia, Depok, West Java, Indonesia. It was part of a more comprehensive study which was aimed to assess the association between dietary intake and hydration status among students in Universitas Indonesia. Data collection was conducted from February 2019 to June 2019. Prior to the data collection, approval from the Ethical committee of faculty of Medicine Universitas Indonesia was obtained (no. 1318/UN2.F1/ETIK/2018).

Sampling

Respondents included in the study met several criteria which were Indonesian citizen, undertaking bachelor degree regular education program, having active student status, and willing to participate in this study. Those, who had serious co-morbidities that could alter the usual dietary intake (e.g. chronic infectious disease, chronic diarrhea, and undergoing any medication or diet therapies) and disability, were excluded from the study. Furthermore, respondents were classified into two groups which are NFP users and non-users group according to their frequency of reading NFP. Those who 'always or most of the times or sometimes' read NFP categorized as NFP users, while those who 'never or rarely' read NFP categorized as NFP non-users.

Sample size needed was calculated using formula for hypothesis test for two population means (two sided test) with 95% confidence level and 80% of power, based on mean difference of diet quality score between NFP users (67.1±12.23) and non-users (60.7±10.11) among university students from previous study (Buyuktuncer *et al.* 2018). After taking into consideration design effect of 1.5 and non-response rate 15%, minimum sample size required were 85 respondents per group or 170 respondents in total for two groups. The respondents were recruited based on non-probability quota sampling. Students who attended the dormitory's canteen, met the criteria of the study, and signed the informed consent were invited directly on voluntary bases until each quota of NFP users and non-users group were filled.

Data collection

An interview using structured questionnaire was conducted to collect data about socioeconomic and demographic characteristics, NFP use, dietary intake, food preparation status and nutritional knowledge. The questionnaire had been pretested previously among 30 subjects who had similar characteristics to the study population. Specifically, nutrition knowledge questionnaire had been tested for its reliability resulted in Cronbach alpha of 0.859. Nutrition knowledge in the current studies encompassed four topics which were dietary recommendation (four questions), source of nutrients (five questions), diet disease relationship (four questions) and understanding of NFP (ten questions). Final score of nutrition knowledge was categorized into two groups either as lower or higher knowledge with cut off of 70 (Iswarawanti 2012).

Food preparation status was defined as any practice of purchasing or cooking food daily by respondents themselves in the past 30 days. Those who had ever done grocery shopping or cooked

food in the past 30 days would be categorized as "yes" while others were "no".

Weight and height measurement was conducted by trained enumerators using standardized protocol (Fahmida & Dillon 2007). Weight was measured using SECA weighing scale 876 to the nearest 0.1 kg, while height was measured using height measuring board wooden (shorr-board) to the nearest 0.1 cm. Weight and height was converted into Body Mass Index (BMI) and classified based on WHO BMI cut offs for Asian population to define the nutritional status of respondents. The categorization was overweight, normal, and underweight for those with BMI ≥23 kg/m^2, 18.5–22.9 kg/m^2 and <18.5 kg/m^2 respectively (WHO 2004).

Dietary quality was measured using a dietary metric, particularly the Diet Quality Index-International (DQI-I) derived from 3x24 hours non-consecutive food recall. Habitual diet of each respondent was calculated using a complex, component specific, and unequal weighted scoring (Miller *et al.* 2020). Briefly, DQI-I scoring system was assigned on four major aspects which were variety (overall food group variety and within group variety for protein), adequacy (vegetable group, grain group, fiber, protein, iron, calcium and vitamin C), moderation (total fat, saturated fat, cholesterol, sodium, and empty calorie foods) and overall balance (macronutrient and fatty acid ratio). Score range of variety, adequacy, moderation and overall balance was respectively 0−20, 0−40, 0−30, 0−10 resulting in maximum score of 100. Higher score indicated higher diet quality and adherence to dietary recommendation (Kim *et al.* 2003).

Prior to data analysis, food database had been developed. Value of energy, carbohydrate, protein, fat, sodium, iron, vitamin C, fiber and calcium were obtained from Food Composition Table (FCT) of Indonesia. If several cooked food, nutrients (e.g. PUFA, MUFA, SFA and cholesterol) and complex dishes were not available in Indonesian FCT, this study applied borrowing technique from other countries' FCTs that provide those information. The similar characteristic to Indonesian circumstances and data completeness became consideration for using other FCTs (MoH RI 2017). In addition, for packaged food and drinks that have NFP attached on it, the nutrients from manufacture's labels were also used.

Data analysis

All analysis were performed using SPSS for Windows version 20.0. Level of significance was set at $p<0.05$. Normality was examined using kolmogorov-smirnov test ($p<0.05$). Since all continuous variables including age and diet quality score had a non-normal distribution, data were presented as median (minimum−maximum value). Meanwhile, categorical data were presented in frequencies and percentage.

A chi square analyses applied to examine the differences of characteristics (gender, education major, mother's education, pocket money, nutritional status, food preparation activity and nutrition knowledge) between NFP users and non-users. The non-parametric mann-whitney test were performed to assess the difference of diet quality score between these two groups as well as age. Furthermore, all variables that had $p<0.25$ in bivariate analysis, were used for multiple linear regressions test to examine the association between NFP use to total diet quality, variety, adequacy, moderation and overall balance score by adjusting the possible confounders particularly for socioeconomic demographic characteristics.

RESULTS AND DISCUSSION

Socioeconomic demographic characteristics

There were 172 respondents participtaed in the study, consisted of 85 NFP non-users and 87 NFP users. Table 1 presented the general characteristics of respondents by NFP use. Age of respondents ranged from 18−21 years old with a median age of 19 years. The majority of respondents were female (59%), had normal nutritional status (53%), taking non-health major program (67%), having mother with more than nine years of education (72.7%), having allocation for food more than median (IDR 900,000 or US$ 62.90) in a month (54.7%), engaged in food preparation activities including cooking and grocery shopping in past 30 days (63.4%) and categorized in lower nutritional knowledge (86%). Almost all characteristics were comparable between NFP users and non-users groups except for the nutrition knowledge, whereas the NFP users were 1.852 times more likely to have higher knowledge than the non-users ($p<0.05$; 95% CI:1.009−3.396).

Table 1. Socioeconomic demographic characteristics of respondents by NFP use

Characteristics	All respondents (n=172)	NFP non users (n=85)	NFP users (n=87)	p
	n (%) or median (min−max)	n (%) or median (min−max)	n (%) or median (min−max)	
Age (years)[a]	19 (18−21)	19 (18−21)	19 (18−21)	0.238[1]
Gender				
Male	70 (41)	36 (42)	34 (39)	0.662[2]
Female	102 (59)	49 (58)	53 (61)	
Education major				
Non-health program	116 (67)	60 (71)	56 (64)	0.384[2]
Health program	56 (33)	25 (29)	31 (36)	
Mother's education				
≤9 schooling years	47 (27.3)	61 (72)	62 (71)	0.942[2]
>9 schooling years	125 (72.7)	24 (28)	25 (29)	
Monthly pocket money for food				
≤IDR 900,000	78 (45.3)	37 (44)	41 (47)	0.636[2]
>IDR 900,000	94 (54.7)	48 (56)	46 (53)	
Nutritional status				
Normal	92 (53)	49 (58)	43 (50)	
Underweight	29 (17)	14 (16)	15 (17)	0.506[2]
Overweight	51 (30)	22 (26)	29 (33)	
Food preparation status				
No	63 (36.6)	30 (35)	33 (38)	0.720[2]
Yes	109 (63.4)	55 (65)	54 (62)	
Nutrition knowledge				
Lower knowledge	148 (86)	80 (94)	68 (78)	0.003[2,*]
Higher knowledge	24 (14)	5 (6)	19 (22)	

NFP: Nutrition Fact Panel use was assessed using a question asking for frequency of reading panel
Response range from 0 to 4; where 0:never and 4:always; Those responding "always"; "most of the time"; and "sometimes" were categorized as users and others as non-users
[a]Median (min-max); [1]Mann-whitney u test analysis; [2]Chisquare test analysis; [*]Significance level at p<0.05
IDR: Indonesian Rupiah

Diet quality of respondents

Total diet quality score and its subcomponents were described in Table 2. No significant different was found in total DQI-I score between the NFP users and non-users. In the two groups, the median score of total diet quality was 44 out of 100. It was categorized low based on the original version of DQI-I which used score ≥60 as the cut off to define good diet quality (Kim *et al.* 2003).

The majority of DQI-I subcategories score had equal median value both in NFP users and

Table 2. Total and component DQI-I scores by NFP use of respondents

Component score of DQI-I	Nutrition fact panel		p[1]
	Non users (n=85)[a]	Users (n=87)[a]	
Variety	9 (3−18)	10 (3−18)	0.061
Overall food group variety	6 (3−15)	9 (3−15)	0.063
Within group variety for protein sources	1 (0−5)	3 (0−5)	0.302
Adequacy	19 (12−30)	19 (12−28)	0.490
Vegetable group	1 (0−1)	1 (0−1)	0.548
Fruit group	1 (0−3)	1 (0−3)	0.609
Grain group	3 (1−5)	3 (1−5)	0.729
Fiber	1 (1−3)	1 (1−3)	0.548
Protein	5 (3−5)	5 (3−5)	0.193
Iron	3 (1−5)	3 (1−5)	0.819
Calcium	3 (1−5)	3 (1−5)	0.003*
Vitamin C	3 (1−5)	1 (1−5)	0.666
Moderation	15 (3−30)	15 (0−24)	0.164
Total fat	3 (0−6)	0 (0−6)	0.517
Saturated fat	0 (0−6)	0 (0−6)	0.570
Cholesterol	6 (0−6)	6 (0-6)	0.468
Sodium	6 (0−6)	6 (0−6)	0.451
Empty calorie foods	3 (0−6)	0 (0−6)	0.052
Overall balance	0 (0−6)	0 (0−6)	0.607
Macronutrient ratio	0 (0−6)	0 (0−6)	0.607
Fatty acid ratio	0 (0−0)	0 (0−0)	1.00
Total DQI-I score	44 (33−63)	44 (29−63)	0.984

DQI-I: Diet Quality Index International

[1]Mann-whitney u analysis test; *Significance level at $p<0.05$; [a]Median (min−max)

non-users group. The median score of DQI-I major subcategories including adequacy, moderation and overbalance respectively were 19, 15 and 0. Unlike the other main subcategories, total variety score in NFP users had slightly higher value than non-users (10 vs. 9). All these scores only achieved ≤50% of perfect score which indicated that the characteristics of diet among university students responding to this study were lack of variety, deficient in favorable nutrients, excessive in unfavorable nutrients and imbalance in proportionality. This typical diet were also found in other studies among Indonesian urban adult population. Those studies consistently revealed

that the median score of total DQI-I, adequacy, moderation and overall balance only fulfill less than half of maximum score (Sartika 2018; Birahmatika 2020).

Association between NFP use and diet quality

The main objective in this study was to assess the association between NFP use and diet quality among university students in Universitas Indonesia. Table 2 showed that there was no statistical difference in total DQI-I score and its subcomponents between NFP users and non-users except for the adequacy score of calcium ($p<0.05$). NFP users compared to non-users had overall higher score. Exploration in the frequency distribution of calcium adequacy score showed that the proportion of NFP non-users got score of 1, 3 and 5 were 15.3%, 76.5% and 8.2% respectively, while NFP users were 4.6%, 75.9% and 19.5% respectively. Study of Graham and Laska (2012) found similar finding. There was slightly (although not significantly) higher intake of calcium and dairy in frequent label readers compared to non-readers.

After controlling for possible confounders including gender and food preparation status, the result revealed that NFP use had no significant contribution to predict total diet quality score (Table 3). It was in contrast to the study conducted by Christoph and An (2018) which found significant association between NFP use and various intake of food groups and nutrients.

NFP use had no association to total adequacy, moderation, and overall balance score as well (Table 4). Nonetheless, it was positively associated to total variety score ($\beta=0.985$; $p<0.05$). It is in line with previous studies among university students that showed NFP users had significant higher consumption of fruit and vegetable compared to non-user (Buyuktuncer *et al.* 2018; Graham & Laska 2012).

Other factors including doing food preparation ($\beta=-1.995$; $p<0.05$) and being a female ($\beta=-1.389$; $p<0.05$) were negatively associated to total variety score (Table 4). Being a female were also contributed to lower total adequacy score ($\beta=-1.513$; $p<0.05$). Those findings were in line with the result from a cross sectional study among university students in Germany. The previous study reported that female students compared to male had significantly lower food group consumption of protein (e.g. poultry, sausages, fish, and cheese) and grain (e.g. pasta and rice). Gradually it might lead to poorer variety and adequacy aspects of diet quality (Hilger *et al.* 2017). Another study discussing dietary intake of Malaysian university students informed that most students (85%) would sometimes prepare their food, yet instant noodles were the most frequent type of food they cooked. Surjadi (2013) through deep interview of several university students in Jakarta also reported that students preferred to cook instant noodle while studying at night since it was more practical and cheaper. Therefore, preparing food activity among university students might contribute to lower variety of food consumed.

The non-significant association between NFP use and diet quality might be explained by some possible rationales. Firstly, NFP users surveyed in this study likely had inadequate understanding to interpret information given on the panel. Consequently, food contained more unfavorable nutrients was selected and gradually affected diet quality. Supporting results on nutrition knowledge questionnaire in this study particularly in the section of understanding NFP showed that less than 25% NFP users could

Table 3. Multiple regression analysis for factors associated to total diet quality score

Independent variables	B	SE(B)	p
Intercept	48.226	1.951	
Food preparation status	-1.839	1.089	0.093
Gender (being a female)	-1.280	1.068	0.233
NFP use	0.118	1.049	0.910

B: Regression coefficient; SE(B): SE of the regression coefficient

R square: 0.026 analyzed with multiple linear regression using enter method; NFP: Nutrition Fact Panel

Table 4. Multiple regression analysis for factors associated to total variety, adequacy, moderation, and overall balance score

Dependent variables	Independent variables	B	SE (B)	p	R square[1]
Total variety score	Food preparation status	-1.995	0.485	0.000*	0.169
	Gender (being a female)	-1.389	0.476	0.004*	
	Age (years)	0.344	0.267	0.2	
	NFP use	0.985	0.469	0.037*	
Total adequacy score	Age (years)	0.393	0.295	0.184	0.095
	Gender (being a female)	-1.513	0.536	0.005*	
	Mother's education	0.371	0.595	0.534	
	Food preparation status	-1.032	0.535	0.055	
	NFP use	0.492	0.519	0.345	
Total moderation score	Age (years)	-0.456	0.419	0.278	0.082
	Gender (being a female)	1.445	0.753	0.057	
	Monthly pocket money for food	-1.471	0.743	0.05	
	Food preparation status	1.028	0.762	0.179	
	NFP use	-1.241	0.735	0.093	
Total overall balance score	Age (years)	-0.218	0.171	0.204	0.019
	Nutritional status	-0.086	0.171	0.616	
	Food preparation status	0.288	0.311	0.356	
	NFP use	-0.122	0.301	0.687	

B: Regression coefficient; SE(B): SE of the regression coefficient; NFP: Nutrition Fact Panel
*Significance level at $p<0.05$; [1]Multiple linear regression analysis using enter method

correctly answered questions asking for a food contained the least calories, least sugar and most sodium from three panels given.

Research suggest that proper understanding and frequent use of nutrition label could assist people to select healthier food and beverage products. Hence, it may improve dietary intake (Lim *et al.* 2015; Liu *et al.* 2015). However, technical and numerical information on nutrition label (e.g. serving size, serving per container and % Recommended Dietary Allowance) were hard to understand by general population (Cowburn & Stockley 2005). In consequence, it might disrupt people to choose healthier food option.

Secondly, respondents participated in current study might had negligence of NFP's information. It was supported by another finding on this study which showed that among students with good understanding, there were possibility to not always use NFP because of time barrier,

price and flavor of food. Consistently, previous studies also implied that nutritional knowledge alone might be not a sufficient reason for label use. Other factors such as foods' price and taste (Mauludyani *et al.* 2021; Talagala & Arambepola 2016), positive attitude to diet (Cowburn & Stockley 2005) and time constraint (Rasberry *et al.* 2007) could influence nutrition label use among adolescent and adult. Finally, another possible consideration to explain the lack of association between NFP use and diet quality may be linked to the overall low diet quality score in total and its components among respondents.

The present study used measures of diet quality which covered various aspects of diet including variety, adequacy, moderation and overall balance to portray the complexity of diet, rather than simply nutrient intake. These measures were developed using non-consecutive 3x24-h recall which increases the validity and reliability of the assessment to represent the habitual diet of the respondents. Nonetheless, the limited FCT available for Indonesia may introduce error in the determination of nutrient content that may lead to misclassification. However, these errors was believed to be at random and did not introduce bias toward the result of the study. Moreover, several effort has been done to minimize the error by using standardized borrowing method considering retention and yield factor (MoH RI 2017).

The study used quota sampling on voluntary bases, and thus may lead to selection bias, where respondents who were keener to their health participated in the study; but the tendency is similar for both groups. The study could only be generalized to university students with similar characteristics to the study subjects. Another limitation include the basis to classify NFP users and non-users which was only from a question asking for frequency reading panel. Future study could add other aspects such as understanding of NFP and attention to specific content reading on panel.

CONCLUSION

This study found that the dietary quality among university students participated in the current study was poor. Although NFP use and total diet quality score had no association, the exploration in each component of diet quality score showed calcium adequacy score was statistically different between NFP users and non-users in which NFP users compared to non-users had overall higher score. In addition, NFP use was also positively associated to total variety score. NFP use may have a role for improving diet quality. Future nutrition intervention could focus to enhance the understanding of technical and numerical information attached on NFP among university students in order to maximize the usage of NFP for selecting healthier food option. Moreover, simplifying NFP's information could also become another strategy to increase the understanding of NFP by general population.

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DECLARATION OF INTERESTS

The authors declare that no conflict of interest with other person or institution.

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Calcium Bioavailability and Serum Calcium Level in Pregnant Rats After Administration of Milk-Based Drinks Containing Lactic Acid Bacteria

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ABSTRACT

This study aims to evaluate calcium bioavailability through serum calcium level in pregnant rats treated with two Milk-Based Drinks (MBD) containing Lactic Acid Bacteria (LAB), i.e *Lactobacillus Casei Shirota Strain* (LcS) drink, and Four Strains Bacteria (FS) (*Lactobacillus rhamnosus, Lactobacillus paracasei, Lactobacillus delbrueckii* subp. *bulgaricus, Streptococcus thermophiles*). It was a completely randomized experimental study using 24 Sprague Dawley rats. Rats were divided into one negative control group with a normal nutritional status (A0) and three undernourished groups. The undernourished groups were divided into one positive control group (A1), one group receiving MBD containing LcS group (A2), and another group receiving MBD containing FS (A3). All rats received high protein diets during pregnancy. The intervention was started in early pregnancy (D0) until 19 days of pregnancy (D19). Blood samples were collected at the D0 and D19. No significant differences in food intake were found among the rats in all groups. Administration of MBD containing LAB in A2 and A3 showed significant ($p<0.05$) increment of calcium bioavailability (30.79±6.88%; 20.44±9.04%). Both MBDs treatment containing LAB showed no significant difference in serum calcium bioavailability ($p>0.05$). The results suggest that MBDs containing LAB are useful in enhancing calcium bioavailability.

Keywords: bioavailability, calcium, rats, serum, undernourished

INTRODUCTION

The first one thousand days of life starting from conception until two years old is a "golden period" where rapid growth of the brain and internal organs happens. Nutritient deficiency within this periode will cause irreversible growth and development disorders (MoH RI 2014). One of many nutritional problems often encountered within one thousand days of life the is Chronic Energy Deficiency (CED) in pregnant women. In 2018, the prevalence of CED in pregnant women was 17.3 % (MoH RI 2018). CED during pregnancy can cause failure in fetal growth or Intrauterine Growth Restriction (IUGR), Low Birth Weight (LBW), birth defects, stunting, even infant death (Wu *et al.* 2012).

Pathophysiology of stunting starts during the fetal life in the womb and it will manifest once the child is reaching the age of two (Millenium Challange Account Indonesia 2014). One of the risk factors for stunting is the low body mass index of mothers in their early pregnancy, which affects linear growth of their babies (Pusparini *et al.* 2016). The prevalence of stunting in children under five years old in Indonesia in 2018 was 30.8% (MoH RI 2018). Childhood stunting can lead to obesity, which increases the risks for degenerative diseases in the future (Black *et al.* 2013). In addition, stunting can cause loss in productivity. Renyoet (2016) stated that the productivity decreases between USD214,850 to 966,930 of Indonesia's total (GDP) because of stunting.

Calcium is an important aspect during pregnancy. Adequate calcium intake during this period supports maternal and fetal bone health and reduce the incidence of hypertension in pregnancy (Camargo *et al.* 2013). Total serum calcium usually falls during pregnancy; thus, mothers need to increase their calcium intake during this period. The high levels of total calcium concentration in cord blood compared to maternal serum, showed that 80% of calcium found in the fetal skeleton at birth crossed the placenta during the third trimester and it is mostly derived from

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dietary absorption of calcium during pregnancy (Kalkwarf & Specker 2002). In Cameroon and Thailand, calcium deficiencies in pregnant women were 94.6% and 55% respectively, meanwhile, in Indonesia such data was not available. However, there was one study conducted by Purnasari *et al.* (2016) showing that 81.2% pregnant women could not meet calcium requirements based on analysis from their daily food intake.

Probiotics or good bacteria provide many health benefits and has become a trend in health and nutrition for gut health in 2021 (KHNI 2021). This benefit is also applied for calcium bioavailability, where probiotics could improve calcium metabolism. Abd El-Gawad *et al.* (2014) showed that yogurt containing *Lactobacillus delbrueckiis* subsp. *bulgaricus, Streptococcus thermophilus, Bifidobacterium longum Bb-46, and Bifidobacterium lactis Bb-12* improve bioavailability of calcium, phosphorus, and zinc in rats. Moreover, Raveschot *et al.* (2020) showed that *Lactobacillus casei, Lactobacillus delbrueckii, Lactobacillus kefiranofaciens, Lactobacillus plantarum, Lactobacillus fermentum*, and *Lactobacillus helveticus* improved absorption of calcium in vitro. There are many researches on probiotics and absorption of calcium which had been done, but so far there are limited reports on the effects of Lactic Acid Bacteria (LAB) consumption on calcium bioavailability and serum calcium both in animal and human with undernourished condition.

In Indonesian market currently there are two popular Milk-Based Drinks (MBDs), the first contain a single species of LAB and the other contain a mixture of four species containing different species of LAB. Considering the positive effect of LAB administration on calcium bioavailability in previous studies and the paucity of data regarding its effect in undernourished pregnant subjects, this study aimed to better understand the effect of MBD with single LAB versus mixtures of LAB in improving calcium bioavalability and serum calcium in undernourished pregnant rats.

METHODS

Design, location, and time

The study was a completely randomized experimental study. The rats were randomly assigned into four gropus. The first group was the negative control group with normal nutritional status (-A0) and the three groups were undernourished groups. The undernourished groups were assigned as the positive control group (-A1) a Milk-Based Drinks (MBD) containing *Lactobacillus Casei Shirota Strain* (LcS) (-A2), and an MBD containing Four Strains Bacteria (FS) (-A3). In the MBD containing LcS and MBD containing FS contain $1x10^9$ CFU/ml and $7.2x10^7$ CFU/ml of total Lactic Acid Bacteria (LAB), respectively. The experiment was conducted in December 2020 to May 2021 at the *Unit Pengelola Hewan Laboratorium* (UPHL), Faculty of Veterinary Medicine, IPB University. The study was approved by Animal Ethics Commission, Faculty of Veterinary Medicine, IPB University No. 025 / KEH / SKE/ 2020. Analysis of serum calcium, feces, and urine ware carried out at the Physiology Laboratory, Faculty of Animal Science, IPB University. Total LAB in the drink products were analyzed at the SEAFAST Center Laboratory, IPB University.

Materials and tools

The main materials used in the study was two MBD brands containing LAB which was obtained from Greenfields Indonesia and Yakult. Six to eight weeks old female (150–250 g) rats were purchased from the Indonesian Rat Company Laboratory (iRATco), Bogor. Male Sprague Dawley (200–250 g) rats were also purchased from iRATco for mating.

Materials and equipment for animal maintenance and treatment were MBD containing LcS (4.3% milk and 76.5% water), MBD containing FS (62.4% milk and 20.8% water), standar rat diet with 18% casein (AIN-93M formulation), low-protein rat diet with 2% casein (Smith *et al.* 2013), high-protein rat diet with 21% casein (AIN93G and Nutrient Requirements of Laboratory Animals 1995), portable digital scales (Sigma), plastic zip, OneMed PCR tube plastic 1.5 ml, individual cage with cage area 33.5x27x12 cm, husk, and drinking bottle sized 250 ml.

Materials used for estrous cycle observation were 0.9% NaCl, 10% giemsa, methanol, distilled water, sensi gloves, cotton buds, object glass, (GEA) and microscope. Materials used for intervention were 1 ml syringe (Terumo) and gavage tube. Materials and equipment used for necropsy and serum collection were 90% alcohol xylazine and ketamine (3:7), surgical

board, surgical blade no.20, operating scissors sharp blunt straight 14 cm, cotton, 1 ml syringe (Terumo), 18 g needle (Terumo), 16 m x 150 mm centrifuge test tube (Kokusan, Japan), gloves (Sensi). Bioavailability of calcium was measured by calculation of the calcium absorption and calcium retention. Calcium absorption, calcium retention, and serum calcium were obtained from analysis of calcium in the urin, feces, and blood serum using spectrophotometry method at wavelength of 422.7 nm.

Procedures

Determination of total LAB in MBD, total LAB, and total coliform bacteria in feses. Total LAB in MBD (from Greenfields Indonesia and Yakult), total LAB, and total coliform bacteria in feses were analyzed using methods according to Bacteriological Analytical Manual (2001). de Man Ragosa and Sharp Agar (MRSA) was used in the total LAB analysis. Meanwhile, Violet Red Bile Lactose Agar (VRBA) was used in the total coliform bacteria analysis. Samples that have been diluted (1 ml) were put into a petri dish, then added with 15–20 ml MRSA or VRBA and shake. Samples were incubated at 37^{o}C in a reverse position for 48 hours. Countable colonies were in the range of 25–250 ml.

Induction of undernutrition. Experimental rats were set and put into separated cages (each cage contained one rat) and exposed to 12-hours light-dark cycle. Induction of undernutrition was done by combination of 50% restrictive diet (Nurliyani *et al.* 2014) and low protein diet with 2% casein (Smith *et al.* 2013). Undernutrition was set at mild condition with 8.3–22.6% body weight loss (Merino-Sanjuan 2014).

Measurement of estrous cycle. The estrous cycle was done by vaginal smears. Sterile cotton-tipped swabs wetted in 0.9% NaCl were gently and quickly introduced into the vaginal orifice, Small cotton swab was wetted with 0.9% NaCl and then swab into the rat's vagina. The swab cotton was carefully rotated (one twist) against the vaginal wall. Rats were not anesthetized during smear collection. The collected sample of vaginal epithelial celss was placed on glass slides, soaked in methanol for five minutes, soaked in 0.1% giemsa for 15 minutes, dried at 37^{o}C and fixed in distilled water for one minute. The cotton swab was then observed under photomicroscope (olympus).

Intervention of MBD containing LAB and measurement of body weight and food intake. The mating process was carried out under a ratio of 1:1 starting from 04.00 p.m to 05.30 a.m. Day 0 of pregnancy was declared if there was sperm from the vaginal smears in the morning after mating. All rats were fed with high-protein diet during pregnancy. Administration of MBD containing LAB was done for A2 and A3 as much as 1.5 ml/day by oral gavage based on stomach capacity of rats of 10 ml/kgBW (McConnell *et al.* 2008). Meanwhile, A0 and A1 groups were given mineral water with the same amount. The MBD containing LcS had 76.9 kcal energy, 1.5 g protein, and 47.7 mg calcium per 100 ml. Meanwhile, 100 ml MBD containing FS had 112 kcal energy, 3.2 g protein, and 80 mg calcium. The female rats delivered their off springs by caesarean surgery within 19 days in accordance with ethical guidelines. Measurement of body weight was done every three days. Meanwhile, the total food intake was measured by food given minus food waste. The food waste was weighted every morning.

Analysis of serum calcium and calcium bioavailability. Blood sampels, urine, and feces were collected at D0 and D19 (before the rats was sacrificed). At the D19, rats were sacrificed for other data collection regarding the fetus sample. The blood, urine, and feces samples were stored in a deep freezer -70^{0}C temporarily until all samples were collected. Analysis of serum calcium, feces, and urine were performed at the end of intervention using spectrophotometer.

Two cc of blood from the tail vein was obtained for the serum collection. The rats were sacrificed at the end of the intervention with a heart pucture under ketamine/xylazine anesthesia and blood was drawn directly from the heart for serum collection (after intervention) and other data collection. The blood was collected in a centrifuge tube and centrifuged at 1,500 rpm for 20 minutes to get the serum. Serum calcium analysis was done using spectrophotometer. As much as 1 ml of serum sample was pipetted into a test tube then added with 4 ml of 5% TCA. The solution wax vortexed and centrifuged for 30 minutes at 3,000 rpm. One ml of the supernatant formed was pipetted into another test tube, then added with 1 ml of 5% Strontium (Sr) solution and 8 ml distilled water. Next, the samples were analyzed using spectrophotometer at a wavelength 422.4

nm. The standard calcium solution used was Calcium Carbonate ($CaCO_3$) (Suhartini 2013).

Measurement of calcium bioavailability was done by calculating calcium absorption and calcium retention. The data on calcium absorption and calcium retention was obtained by calculating calcium intake from the diet (including calcium content of MBD) and analyzing calcium in urine and feces. Urine and feces of the rats were collected and put into a 100 ml Erlenmeyer two days before the intervention and two days before the rats were sacrificed. As much as 5 ml HNO_3 was added into the sample and then the samples were put in the acid chamber. After one hour, the samples were heated on a hot plate at low temperature for 4–6 hours in the acid chamber. After keeping it for overnight, 0.4 ml H_2SO_4 was added into the samples and they were heated on a hot plate again to concentrate the solution. Then, 2–3 drops of a mixed solution of $HClO_4$:HNO_3 (2:1) was added into the samples. The heating was continued until the color changed from brown to dark yellow, and then back to light yellow (approx. one hour). After the color change had been seen, the heating process was continued for 10–15 minutes. The samples were cooled and added with 2 ml distilled water and 0.6 ml HCI and then heated again so that the samples dissolved. The samples were next put into a 100 ml volumetric flask. If there was a precipitate, it was filtered with a glass wool. The results of wet ashing were analyzed using spectrophotometry to determine the calcium concentration at of 422.4 nm. The bioavailability of calcium was measured by calculating the calcium absorption and calcium retention.

Data analysis

Data on body weight, food intake, serum calcium, and calcium bioavailability were processed using Ms.Excel 2013. The results were presented as mean±Standard Deviation. Statistical significance was evaluated using one-way ANOVA and continued with Duncan Multiple Range Test (DMRT) at 5% (0.05). The statistical analysis was done using SPSS software version 16.0.

RESULTS AND DISCUSSION

Effects of milk-based drink containing lactic acid bacteria on food intake and body weight of rats

Data of food intake of rats was collected by weighing the leftover of feed every day. Table 1 shows the food intake of rats during pregnancy. Table 1 show that there was no significant difference in food, energy, and protein intake between all groups. Data of rats' body weight is shown in Table 2.

Table 1 shows that there was no significant difference in body weight loss because of induction undernutrition among A1, A2, and A3. This due to the same decrease in food intake. Weight loss in a short period of time can be caused by treatment of decreasing food intake (Estrela *et al.* 2014). Food restriction by 50% and low protein diet (2% casein) can lead to loss of skeletal muscle, which is very sensitive to protein deficiency because it is a protein reservoir to the organism. Protein deficiency makes depletion of the tissue, leading to significant muscle loss and consequently decreased body mass. Thus, mothers who are undernutrition have higer risk to deliver Low Birth Weight (LBW) babies (Yongki *et al.* 2014).

MBD containing *Lactobacillus casei shirota strain* (LcS) drink was given to A2, and MBD containing four strains bacteria (FS) (*Lactobacillus rhamnosus, Lactobacillus*

Table 1. Food and nutrient intake of rats during pregnancy

Group	Food intake (g)	Energy (kcal)	Protein (g)
A0	18.05±2.53	63±9.10	4.12±0.58
A1	20.88±1.05	73±4.00	4.76±0.24
A2	21.08±2.05	75±7.20	4.83±0.47
A3	21.42±1.68	76±5.90	4.92±0.00
p[1]	0.090	0.069	0.078

[1]One way-ANOVA

Table 2. Effect induction of undernutrition on rats weight loss and effects of MBD containing LAB on rats body weight during pregnancy (day 0, day 19, and increment body weight)

Group	Weight loss (%)	Day 0 (D0)	Day 19 (D19)	Increment (D19–D0)
A0	-	153.67±19.37[a]	224.33±23.29	70.67±5.43[a,b]
A1	10.15±1.69	201.67±17.50[b]	276.33±15.57	74.67±2.31[b]
A2	12.37±1.82	182.50± 21.21[a,b]	245.00±30.01	62.75±9.74[a]
A3	12.69±2.76	172.00±19.80[a,b]	253.00±27.62	81.00±8.54[b]
p[1]	0.322[*]	0.026[*]	0.069	0.031[*]

[1]One-way ANOVA analysis ; [*]Significant at p<0.05
MBD: Milk-Based Drinks ; LAB: Lactic Acid Bacteria

paracasei, Lactobacillus delbrueckii subp. bulgaricus, Streptococcus thermophiles) was given to A3. Statistical analysis showed that A2 (62.75±9.74 g) had significantly lower weight gain compared to A1 (74.67±2.31 g) and A3 (81.00±8.54 g). The gross body weight gain of rats in positive control group (A0) at D19 of pregnancy was ±45.9% of initial body weight. Study conducted by Rahayu (2020) showed that gross body weight gain of rats was up to 72.83% at partus. In this study, the smaller weight gain in A2 at 34% of initial body weight can be associated with the administration of MBD containing LcS. Another animal study by Karimi *et al.* (2015) also showed the same results where the administration of LcS to rats which were given high-fat diet had caused a significantly lower final body weight compared to rats which were fed with a high-fat diet only.

On the other hand, an animal study conducted by Benakriche *et al.* (2014) showed that in malnourished rats treated with *Lactobacillus acidophilus, Lactobacillus rhamnosus, Bifidobacterium lactis, Bifidobacterium longum, Bifidobacterium bifidum, Streptococcus thermophiles,* and fructooligosaccharides at 0.5 mg/g of body weight/day managed to improve the rats' intestinal atrophy that was caused by malnutrition; thus, the rats' body weight increased from 123 g±9.9 g to 181.08±9.9 g. However, the weight gain of A3 (81.00±8.54 g) which was given FS in this study was not significantly different from the control groups (A0 and A1). This might be due to the dose of MBD containing FS that was not enough to increase the rats' body weight during pregnancy higher than the standard high protein diet given to the control groups. It was noted that beverage product for A3 contained $7.2x10^7$ CFU/ml total LAB. Based on the World Health Organization, the recommended dose of probiotic to provide health benefits is between 10^8–10^9 CFU/ml.

Some probiotics can help increase body weight while other probiotics work adversely. Several mechanisms underlie how bacteria strains elicit their function. Oral administration of probiotics increases the activity of the sympathetic nervous system in white and brown adipose tissues. Meanwhile, intragastric administration of probiotics increases lipolysis in white adipose tissue and thermogenesis in brown adipose tissue (Tanida *et al.* 2008). This process facilitates thermogenic and lipolytic responses via stimulation of the sympathetic nervous system, which leads to weight reduction. Another mechanism underlying how probiotic induce weight reduction is the bacteria's metabolite known as Short Chain Fatty Acids (SCFA), especially butyrate. Butyrate plays as the main regulator of energy production and mitochondrial function. It induces gene expression of peroxixome proliferator-activated receptor-gamma coactivator-1 alpha (PGC-1α) in the muscle and brown adipose tissue where PGC-1α is involved in thermogenesis and glucose metabolism process. Therefore, it affects body weight loss (Gao *et al.* 2009).

Probiotics administration also benefits the host through vitamin synthesis and amino acids release. The MDB yogurt given to A3 contained 62.4 % milk. There are activities from *Lactobacillus delbrueckii subsp.* bulgaricus and *Streptococcus thermophiles* that allow the pre-digestion of milk protein. Those processes help

produce more lactose which can be digested and it is more tolerable than liquid milk (Savaiano 2014). This mechanism helps increase the body weight of experimental rats. Dainese–Plichonet *et al.* (2014) also showed that the consumption of yoghurt and cheese was recommended as part of diet to help people with lactose intolerance to still be able to gain benefits from dairy products.

Effects of milk-based drink containing lactic acid bacteria on calcium bioavailability

Calcium from food is usually unable to be 100% absorbed by the body because it depends on biological availability or bioavailability of it. Moreover, positive calcium balance is required, especially during growth, pregnancy, and lactation period. Average values from absorption, retention, bioavailability, and increment of bioavailability of calcium in pregnant rats before and after the intervention are displayed in the Table 3.

Based on the baseline data shown from Table 3, it showed that calcium absorbtion, calcium retention, and calcium bioavailability of rats with undernutrition induction (A1, A2, and A3) were significantly lower compared to rats with normal weight (A0). Table 3 shows absorption, retention, and calcium bioavailability in A1, A2, and A3 that were significantly lower than A0. This might be due to differences in calcium content for standard and low-protein diet. The low-protein diet for induction of undernutrition, had 0.88 g calcium per 100 g while the standard diet had 0.90 g calcium per 100 g. In addition, the amount of food was also restricted for A1, A2, and A3.

With induction of undernutrition can interfere calcium absorption. Protein is the largest part in human body after water. Enzymes, hormones, nutritional carriers, blood, and intracellular matrix are made from protein. Protein deficiency causes disturbances in nutrients absorption and transport (Nyaradi *et al.* 2013). In this study, protein deficiency could possibly affect calcium bioavailability.

Administration of MBD containing LAB led to significant increase of calcium bioavailability ($p<0.05$) in all groups (A2 and A3). The Duncan test showed that in calcium bioavailability in A2 (30.79±6.88%) was significantly higher compared to A0 (1.01±4.46%) and A3 (20.44±9.04%). Similar results were also reported by Abd El-Gawad *et al.* (2014) who showed that administration of *Bifidobacterium lactis* and *Bifidobacterium longum probiotics* in rats for 45 days resulted in higher calcium absorption (84% for *Bifidobacterium lactis* and 83.4% for *Bifidobacterium longum*) than the control group

Table 3. Effects MBD containing LAB on the absorbtion, retention, bioavailability of calcium

Parameters	Groups				p[1]
	A0	A1	A2	A3	
Calcium absorption ability (%)					
Before	68.1±11.04b	22.87±5.73[a]	37.35±13.18a	30.13±13.14[a]	0.000*
After	51.08±25.90	71.77±25.36	81.05±2.74	74.40±29.83	0.245
Calcium retention (%)					
Before	62.42±11.54[b]	13.3±4.76[a]	25.12±10.10[a]	23.57±12.19[a]	0.000*
After	49.32±25.28	68.57±27.89	78.45±3.18	71.50±29.10	0.276
Calcium bioavailibility (%)					
Before	91.35±310[c]	57.37±10.02[a]	66.03±6.72[a,b]	75.47±9.39[b]	0.000*
After	92.35±6.02	93.85±7.15	96.79±1.34	95.90±0.99	0.535
Increment bioavailability (%)	1.01±4.46[a]	36.48±14.48[c]	30.79±6.88[b,c]	20.44±9.04[b]	0.000*

[1]ANOVA one-way analysis; MBD: Milk-Based Drinks ; LAB: Lactic Acid Bacteria

(66%). The groups of rats which were given probiotics had an increase in calcium absorption by 24.7%– 26.6%. Administration of probiotics is proven to increase the crypth depth in the large intestine and it has the ability to lower pH due to increasing Short Chain Fatty Acids (SCFA) production in the large intestine. Decreasing pH causes increased calcium absorption (Abd El-Gawad *et al.* 2014).

In vivo study conducted by Raveschot *et al.* (2020) reported that probiotics *Lactobacillus casei, Lactobacillus kefiranofaciens, Lactobacillus plantarum, Lactobacillus helveticus, and Lactobacillus delbrueckii* improved calcium transport and uptake. Moreover, *L.casei9b, L. kefiranofaciens15b,* and *L. helveticus49d* increased total calcium transport by increasing calcium solubility. Meanwhile, *L. delbrueckii50b* strain increase calcium transport through paracellular pathway by upregulating the cld-2 gene where claudin-2 functions as a transport channel for ions and water (Lu *et al.* 2013). The use of LAB *L. delbrueckii* and *L. casei* in this study was assumed to have a role in calcium absorption by increasing calcium solubility and upregulating genes that are involved in paracellular calcium transport through intestinal cells.

Milk also affects calcium bioavailability. The MBD given to A2 contained 4.3% milk while MBD given to A3 had higher milk content (62.4%). Milk and dairy products are the main sources of calcium because it has an anti-resorptive effect and can help to prevent bone loss. Besides, the presence of phosphorus in milk and dairy products can also bind calcium to form the hydroxyapatite mineral that are able to increase bone density (Heaney 2002).

Effects of milk-based drink containing lactic acid bacteria on serum calcium

Table 4 shows mean serum calcium in all groups before and after the intervention. The ANOVA test results showed that the induction of undernutrition had a significant effect on serum calcium level. The groups A1, A2, and A3 had significantly lower level of serum calcium than the control group (A0), whereas A3 had the lowest level of serum calcium when compared to all the other groups

These findings was supported by a previous study conducted by Aminah *et al.* (2017) who concluded that normal level of serum calcium in two months Sprague Dawley rats was 11.36±0.46 mg/dl. In this research, the A1, A2, and A3 had a lower serum calcium as much as 17.08%, 19.45%, and 31.55%, respectively when compared to the control group (A0). This might be due to the induction of undernutrition. Study by Solang (2017) showed that induction of undernutrition in rats by administering an 8.46% protein diet for eight weeks showed a decrease in serum calcium level by 31.4% compared to the control group given 13–15% protein for 8 weeks. Moreover, calcium level in the control group was 11.8±0.20 mg/dl (Solang 2017).

The decrease in serum calcium in undernourished group is presumably caused by low protein diet which led to a decrease in the availability of proteins in the body, such as the availability of albumin. Albumin plays an important role in transporting calcium, which means that there is a relationship between calcium absorption and protein intake. Therefore, low-protein diet causes lower calcium absorption; hence, the level of calcium in the blood decreases (Solang 2017). Moreover, induction

Table 4. The average serum calcium levels in rats before and after intervention

Groups	Before (mg/dl)	After (mg/dl)	Δ (mg/dl)	p[1]
A0	10.97±0.97[c]	9.59±1.85[a]	-1.37±2.51[a]	0.239
A1	9.09±0.53[b]	9.01±0.47[a]	-0.08±0.27[a]	0.642
A2	8.83±0.74[a,b]	8.75±0.66[a]	-0.09±0.96[a]	0.867
A3	7.51±0.89[a]	9.49±1.81[a]	1.99±1.93[a]	0.216
p[2]	p[2]=0.000*	p[2]=0.804	p[2]=0.146	

*[1]Paired t-tests analysis before and after intervention; *[2]One-way ANOVA analysis

of undernutrition resulted in lower *Lactobacilli* which caused increased intestinal pH. Eventually, increased intestinal pH caused decreased calcium absorption.

Administration of MBD containing LAB did not significantly affect serum calcium level during pregnancy. This is in line with the results of others studies elaborated before who found that administration of MBD containing LAB decreased intestinal pH and increased calcium absorption. This phenomenol implies that the human body keeps the homeostatic level of serum calcium and it is not affected by calcium absorption. An animal study conducted by Gonen *et al.* (2005) showed that serum calcium level of pregnant rats was 7.86±1.3 mg/dl, which was lower than the results found in this present study. This difference might be caused by the fact that Gonen *et al.* (2005) had no intervention in terms of calcium intake. Descreasing serum calcium in late pregnancy period is associated with decreasing calcium ionization. This might be related to an increase in fetal requirement, and a decrease in serum albumin concentration due to hemodilution, as well as increase in urinary calcium excretion (Prentice 2000). This research showed that although the administration of MBD containing LAB did not provide a significant difference in increasing serum calcium, it was able to maintain the calcium level within the normal range during pregnancy.

CONCLUTION

Intervention of 1.5 ml/day MBD containing LAB during 19 days of pregnancy ($7.2x10^7$ CFU/ml total LAB for A3 and $1.2x10^9$ CFU/ml total LAB for A2) showed significant effect on weight gain and increased calcium bioavailability in undernourished pregnant rats. Administration of MBD containing FS (A3) was associated with higher body weight increase as compared to standard feed. Both MBD containing LcS (A2) and FS (A3) were associated with higher calcium bioavailability. However, MBD containing LAB did not significantly affect the serum calcium in undernourished pregnant rats.

Further research is needed on the development of food product containing a mixture of these LAB to improve calcium bioavailability and serum calcium of undernourished pregnant subjects. The appropriate dosage of the LAB to be administered to undernourished pregnant subjects also needs to be identified.

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DECLARATION OF INTERESTS

The authors declare no conflict of interest.

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Diet Quality and Growth Status of Children Aged Two to Six Years at Tuba Island, Langkawi, Malaysia

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ABSTRACT

This study aimed to determine the prevalence of undernutrition, diet quality and its associated factors among children aged two to six years at Tuba Island. A total of 67 children paired with their mother or caregiver were participated in this study. A questionnaire containing socio-demographic characteristics, anthropometric measurements and two days of 24-hours dietary recall was administered. The Malaysian Healthy Eating Index (HEI) was used to assess the children's diet quality. The prevalence of underweight, stunting and wasting were 22.4%, 20.8% and 17.9%, respectively. Mean total HEI score was 37.1±6.3, which indicate poor diet. The children had low mean scores for vegetables, fruits, milk and dairy products as well as legumes. For socio-demographic factors, number of children per household was associated with diet quality. This study points out that overall low dietary score among these children and their diet quality had no association with their growth status. The low diet quality was prevalent; thus it did not contribute to differences in growth status among these children. Therefore, it is suggested further intervention should be taken to improve the nutrition quality of these children.

Keywords: child undernutrition, diet quality, growth status, healthy eating index

INTRODUCTION

According to World Health Organization (WHO), malnutrition refers to inadequacy, excesses, or imbalances in energy and nutrient intake (WHO 2021). Undernutrition can be categorized into three, namely underweight, stunting and wasting. According to the United Nation Children's Fund (UNICEF), undernutrition has been dubbed as the 'silent emergency'. It has posed a great health risk to women and children worldwide (UNICEF 2021). One out of four children under five years of age in developing countries is undernourished, irrespective of the worldwide decline in underweight prevalence from 1990 to 2010 (United Nation 2011). Malaysia is one of South-East Asia countries with an upper-middle income. However, childhood malnutrition persists in the country and has become a general concern in Malaysia, particularly in rural communities (Wong *et al.* 2014). In a nutrition survey at national level, the prevalence rates of stunting and thinness are 8.3%–8.8% and 5.2%–6% respectively among urban and rural children aged six months to 12 years (Poh *et al.* 2013). In addition, a study in a selected kindergarten from urban and rural areas in Selangor found that the rates of stunting and wasting in children from rural areas were higher with 28.6% and 31% compared to children living in an urban region with 15% and 22% respectively (Aziz & Devi 2012). Same findings were found among indigenous population across states in Malaysia ranging from 35 % to 76 % of indigenous children under five years old were suffering from undernutrition, where stunting was more prevalent than underweight (Chua *et al.* 2012; Geik *et al.* 2016; Murtaza *et al.* 2018; Yen *et al.* 2018).

An individual's dietary intake determines the quality of a diet; thus, it is important to adequately consume food with high quality and quantity of various nutrients in order for the body to sufficiently absorb macronutrients and

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micronutrients (Ihab 2015). Adequate intake during the first two years of life plays a vital role for the nutritional status of children in the future, as it can become a risk factor for stunting (UNICEF 2013). Individual, particularly children suffering from undernutrition may susceptible to illness, delayed mental and motor development as well as other health risks that may take a toll on their health and resulting in adverse effect beyond their childhood (WHO 2019). Good quality diet may represent adequate nutrient intake that children need to improve their growth and development in physical, cognitive and psychosocial terms (Cheng & Buyken 2013). In rural areas, malnourished children are vulnerable to insufficient food intakes, which will lead to macronutrient and micronutrient deficiencies. This deficiency reflects their poor diet quality (Cheng & Buyken 2013). Malaysian Healthy Eating Index (MHEI) has become one of the recognizable tools to assess diet quality in Malaysia reflecting the local context, thus was used in this study.

Tuba Island is situated in Langkawi and it is smaller than the main island thus having smaller geographical areas. Tuba Island is considered a rural area as it is still constrained by remoteness and under-development and limited livelihood opportunities, therefore it was chosen as the study location. According to previous research, geographical differences affect the access of food which are linked to economic barriers to acquiring healthy food options (Vilar-Compte *et al.* 2014). To date, communities living on the island still maintain traditional occupation and culture. The village in Tuba Island was known as a traditional fishing village since their main source of income was from working as fishermen, which may contribute to their low socio-economic status. Thus, the objectives of this study were: (1) to determine the prevalence of undernutrition among children aged two to six years; (2) to assess diet quality status among children and its association with socio-demographic variables and (3) to investigate the association between diet quality and growth status of children in Tuba Island.

METHODS

Design, location, and time

A cross-sectional study was conducted in Tuba Island, Langkawi, located off the main island. It is situated five kilometers southwest of Kuah Jetty, Langkawi. This study was conducted for two weeks in August 2020.

Sampling

The convenience sampling method was used to search for potential subjects. The respondents were approached by researchers by visiting from home to home during data collection. According to the local authority, the number of children and women within the inclusion criteria was limited and therefore, convenience sampling was used. The sample size for this study was calculated using the formula developed by Cochran (Cochran 1963). The estimated proportion is 0.807 based on the prevalence of poor diet quality in Kuala Lumpur (Rezali *et al.* 2015) which is 80.7% desired confidence level of 95% and desired precision of 0.1. Therefore, the required sample size was 59. The prevalence of poor diet quality from Kuala Lumpur is chosen as it has higher prevalence compared to other study.

Attrition rate of 20 % was used to increase the number of respondents participated in this study, therefore, the final total sample size obtained was (n=71). Children aged two to six years were paired with their mother or caregiver. In a household where a mother had more than one child age between two and six years, the youngest one was selected. In this study, children were excluded if they had physical impairment, intellectual development delays, and chronic disease that can affect child's growth includes congenital and acquired diseases. Intellectual developmental delays were observed by the researchers through the children's ability to know full name, gender and understand basic instructions.

Data collection

The data was collected in the participants' house and performed in August 2020. A total of 67 children and care givers dyads were qualified to participate after screening for inclusion and exclusion criteria. Ten children were ineligible to join the study since their age did not meet the requirement. Before data collection process, the research protocol was explained to the caregivers of these children. If they agreed to participate, verbal and written informed consent were signed. Safety measures during Covid-19 pandemic were implemented, where researcher was obligated to wear face masks and gloves as well as

sanitized before and after data collection process. Respondents also were sanitized and their body temperature were checked. Social distancing (one meter) was applied during interview and anthropometric measurements. The session was limited to only 15 to 20 minutes. The application of ethical review was submitted and approved by Research Ethics Committee (REC) Universiti Teknologi MARA (UiTM) with reference number REC/05/2021 (UG/MR/421).

Household demographic and socio-economic status. A face-to-face interview was conducted with the caregiver using a structured questionnaire. The questionnaire consists of 11 items, including the age of mother, marital status, educational level of father and mother, household income, number of persons contribute to household income, household size category, employment status of father and mother, number of children per household and household food expenditure

Anthropometric measurements. The subject's weight was measured using SECA digital weighing scales (to the nearest 0.1 kg) while the height was measured using non-stretchable measuring tape. In order to increase the accuracy of the height measurement, the subjects were told to remove shoes, hats and any other head wears and feet need to be flat on the floor while standing straight with the back against the wall. The date of birth of subjects were obtained from their birth certificates. The age, weight and height of the children were translated into three indices; weight-for-age, height-for-age and weight-for-height, where the z scores were obtained based on the WHO Child Growth Standard median. Underweight was defined as weight-for-age less than -2 SD, stunting was defined as height-for-age less than -2 SD and similarly, wasting was defined as weight-for-height less than <-2 SD.

Dietary intake assessment. Dietary intake of the children was obtained by interviewing the mother or the caregiver using the 24-hours diet recall on one day from weekday and one on weekend. The mothers were asked to remember the foods and beverages consumed by their children in the last 24-hours with the estimation of serving sizes using household measures such as teaspoons, tablespoons, cups, glass, bowls, saucer, etc. along with the Food Atlas: Size Portion and Exchange to achieve a more reliable estimate of portion sizes (Suzana *et al.* 2015). The data obtained were analyzed using Nutritionist Pro Software and compared to the Recommended Nutrient Intake (RNI) for Malaysia (NCFFN 2017).

Malaysian Healthy Eating Index (MHEI). Malaysian Healthy Eating Index was developed by Lee *et al.* (2011) and was used to assess the diet quality of an individual (Lee *et al.* 2011). MHEI consist of seven components of food groups (grains and cereals, vegetables, fruit, meat, poultry and eggs, fish and seafood, legumes, milk and milk products) and two components of nutrients (percentage of energy intake from fat and sodium), with a total of nine components. The calculation scoring component was based on the recommended serving size in the Malaysian Dietary Guidelines for Children and Adolescent (MDG) (Majlis Keselamatan Makanan dan Pemakanan Kebangsaan & Malaysia 2013). The score for each HEI component is computed using the formula:

$$\frac{actual\ serving\ consumed\ based\ on\ respondent's\ diet\ recall}{recommended\ serving\ size\ based\ on\ MDG} \times 10$$

Formulas for composite score:

$$\frac{total\ score\ obtained\ from\ 9\ components}{maximum\ score\ of\ 90} \times 100\%$$

The total HEI score will be obtained by summing up the scores for each element. A composite score above 80% indicated a good diet, a score between 51% to 80% indicated needs of improvement and a score of less than 51% indicated poor diet (Pei *et al.* 2018).

Data analysis

The data collected was analyzed using Statistical Package for Social Sciences (SPSS) version 25.0. Descriptive statistics were reported as mean±standard deviation, frequencies and percentages. Data for anthropometric measurements were analyzed using WHO AnthroPlus. To generate value for weight-for-age z scores (WAZ), height-for-age z scores (HAZ) and weight-for-height z scores (BAZ), the birth date, height and weight measurements of the children were inserted into the software. The association between socio-demographic variables with diet quality were determined by Fisher Exact test. In contrast, the Pearson Correlation test was used to determine the correlation between the children's HEI score and growth status, the children's HEI score and growth status. P-value of less than 0.05 was considered statistically significant.

RESULTS AND DISCUSSION

Table 1 shows the socio-demographic characteristics of 67 household participated in the study. Most of the parents, father and mother had secondary level education with the proportion of 62.7% and 73.1% respectively. For monthly household income, nearly one third (29.9%) of the family has an income less than RM908 (USD218) or considered as below the poverty line. As for the parents' employment status, most fathers were self-employed with a proportion of 58.2%, who mostly worked as fishermen, while most mothers were housewives, 74.6%.

Table 1. Socio-demographic characteristics of respondent

Respondent characteristics	Number (n=67)	Percentage (%)	Mean±SD
Age of mother (year)			34.9±6.8
Marital status			
Married	62	92.5	
Widowed/divorced	5	7.5	
Education level of father			
Primary education	20	29.9	
Secondary education	42	62.7	
Tertiary education	2	3.0	
Education level of mother			
Primary education	16	23.9	
Secondary education	49	73.1	
Tertiary education	2	3.0	
Household income (RM)			
≤RM908 (USD218)	20	29.9	
RM909–RM2,208 (USD219–USD532)	42	62.7	
≥RM2,209 (USD532)	5	7.5	
Number of persons contribute to household income			
None	3	4.5	
Only	48	71.6	
More than one	16	23.9	
Household size category			
≤5 members	34	50.7	
6–8 members	29	43.3	
≥9 members	4	6.0	
Employment status of father			
Government/private sector	25	37.3	
Self-employed	39	58.2	
Employment status of mother			
Government/private sector	8	11.9	
Self-employed	9	13.4	
Housewife	50	74.6	
Number of children per household			
≤3 children	43	64.2	
4–6 children	23	34.3	
≥6 children	1	1.5	
Household food expenditure (RM)			
≤RM500 (≤USD120)	37	55.2	
RM501–RM749.99 (USD120–USD180)	22	32.8	
≥RM750 (≥USD180)	8	11.9	

RM: Ringgit Malaysia; USD: United States Dollar

Of the 67 children participated in this study, 55.2% were male and 44.8% were female. In this study, more than half of the children had normal growth. Twenty-two point four percent were underweight, 20.8% were stunted and 17.9% were wasted (Table 2).

Table 3 displays the average score of each component and total HEI score. The mean HEI score was 37.1±6.3 which indicates that children in Tuba Island were overall had poor diet quality. The low mean scores for the components of the seven food groups, were specifically for

Table 2. Anthropometric characteristic of children

Variables	Frequency (n=67)	Percentage (%)	Mean±SD
Weight (kg)			15.5±4.1
Height (cm)			101.2±13.5
Age (months)			52.2±17.7
24–36	20	29.9	
37–72	47	70.2	
Gender			
Male	37	55.2	
Female	30	44.8	
Weight-for-age (WAZ)			-0.9±1.6
Severe underweight	4	6.0	
Moderate underweight	11	16.4	
Normal	49	73.1	
Overweight	3	4.5	
Height-for-age (HAZ)			
Severe stunting	7	10.4	-0.8±1.9
Moderate stunting	7	10.4	
Normal	52	77.6	
High	1	1.5	
Weight-for-height (BAZ)			-0.5±2.0
Severe wasting	4	6.0	
Moderate wasting	8	11.9	
Normal	41	61.2	
Possible risk of overweight	9	13.4	
Overweight	1	1.5	
Obese	4	6.0	

Table 3. The average score of each component and total HEI score

HEI components	Score range	Criteria for minimum score 0 (serving/day)	Criteria for maximum score 10 (serving/day)	Average score (Mean±SD)
Food groups				
Grains and cereals	0–10	0	2–3	8.1±1.8
Vegetables	0–10	0	2	1.8±1.8
Fruits	0–10	0	2	0.6±1.1
Meat, poultry and eggs	0–10	0	½	7.5±2.7
Fish and seafood	0–10	0	½–1	6.6±2.9
Legumes	0–10	0	½	0.0±0.0
Milk and dairy products	0–10	0	2	3.0±2.3
Nutrients				
Total fat	0–10	≥30% energy from fat	≤25% energy from fat	1.0±2.5
Sodium	0–10	≥1,900 mg	≤1,200 mg	1.0±2.5
Total HEI score	0–100	-	-	37.1±6.3

HEI: Healthy Eating Index

vegetables (1.8), fruits (0.6), milk and milk products (3.0) and legumes (0.0). This suggests a low level of intake of these food groups as opposed to the dietary guidelines. However, the subjects obtained high scores of 8.1±1.8, 7.5±2.7 and 6.6±2.9 for grains and cereals, meat, poultry, eggs, fish, and seafood components. The children obtained low mean scores for percentages of energy from fat with 1.0±2.5 for nutrient components.

Fisher Exact Test result indicates an association between diet quality status and the number of children per household (p=0.044). Families with less than three children in a household had higher percentages of children with poor diet (63.5%) than families with more than three children (Table 4).

Table 5 shows the correlation between HEI score and growth status indices which is weight-for-age (WAZ), height-for-age (HAZ) and weight-for-height (BAZ). The results revealed that there was a non-significant relationship in those three growth status indices of WAZ, HAZ and BAZ with HEI score. Thus, this finding

Table 4. Socio-demographic characteristics by diet quality status of the children aged two to six years

Variables	Need improvement (n=4)	Poor (n=63)	p
	n (%)	n (%)	
The educational level of the father			0.546
Primary	0 (0.0)	21(31.7)	
Secondary/tertiary	3 (75.0)	41 (65.1)	
The educational level of the mother			1.000
Primary	1 (25.0)	15 (23.8)	
Secondary/tertiary	3 (75.0)	48 (. 76.2)	
Household income (RM)			1.000
<RM98 (<USD218)	1 (25.0)	19 (30.2)	
RM909–RM2,208 (USD219–USD532)	3 (75.0)	39 (61.9)	
>RM2,209 (>USD532)	0 (0.0)	5 (7.9)	
No. of persons contribute to household income			1.000
None	0 (0.0)	4 (6.3)	
Only	3 (75.0)	44 (69.8)	
More than one	1 (25.0)	15 (23.8)	
Household size category			0.157
≤5 members	4 (100.0)	30 (47.6)	
6–8 members	0 (0.0)	29 (46.0)	
≥9 members	0 (0.0)	4 (6.3)	
Employment status of the father			0.055
Government/private sector	3 (75.0)	22 (34.9)	
Self-employed	0 (0.0)	39 (61.9)	
Employment status of the mother	0 (0.0)	8 (12.7)	
Government/private sector			0.700
Self-employed	1 (25.0)	8 (12.7)	
Housewife	3 (75.0)	47 (74.6)	
Number of children per household			0.044*
≤3 children	3 (75.0)	40 (63.5)	
4–-6 children	1 (25.0)	23 (36.5)	
Household food expenditure (RM)			0.063
≤RM500 (≤USD120)	2 (50.0)	35 (55.6)	
RM501–RM749.99 (USD120–USD180)	2 (50.0)	22 (34.9)	
≥RM750 (≥USD180)	0 (0.0)	6 (9.5)	

Fisher's exact test was used since cells have an expected count of less than 5; RM: Ringgit Malaysia; USD: United States Dollar.

Table 5. Correlation between HEI score and growth status

Variables	HEI score	p
	r	
Weight-for-age (WAZ)	-0.035	0.778
Height-for-age (HAZ)	-0.008	0.947
Weight-for-height (BAZ)	-0.072	0.562

Correlation is significant at the 0.05 level (2-tailed)
HEI: Healthy Eating Index

suggested that their diet quality cannot indicate the growth status of the children due to the large majority had poor diet.

This study examined the prevalence of undernutrition of children in Tuba Island, Langkawi. The overall prevalence of undernutrition in this study are 22.4%, 20.8% and 17.9% respectively for underweight, stunting and wasting. In Indonesia, the percentage of stunting, underweight and wasting were higher in rural compared to urban areas with 39.2%, 28.9% and 6.0% respectively which is slightly higher than findings in this study (Sandjaja *et al.* 2013). In another study in Philippines, the rate of underweight, stunting and wasting was 26.4%, 37.7% and 11.2%, which is slightly higher than this study findings. Children in a Household Headed by Fisherfolks (HHF) had the highest prevalence of undernutrition among other occupational groups, including households headed by forestry and related workers, compared to other occupational groups (Capanzana *et al.* 2018). Tuba Island is considered a rural region due to its low population density and traditional fishing villages. Thus, as most of the children living in Tuba Island come from families of fishermen, the result was consistent with this study. In comparison with a study in other rural area in Malaysia, the prevalence of wasting and stunting among children in fishermen community in Terengganu were 36.6% and 6.7% respectively (Bahtiar *et al.* 2021).

A healthy and balanced diet is crucial for children's development and growth. Despite the importance of having a good diet quality, most of the children living in Tuba Island had poor diet quality. Most children did not comply with the recommended intake of fruits, vegetables, milk and dairy products and legumes, as shown by the low average score for these components. However, most of the children exceeded both the recommended sodium intake and the percentage of energy intake from fat. This result partially aligns with those of Koo *et al.* (2016) who reported that the dietary intake of Malaysian children for six food groups, including cereals/ grains, fruits, vegetables, legumes, fish and dairy products, was below the recommended levels in the Malaysian Dietary Guidelines (MDG) (Koo *et al.* 2016). The low consumption of dairy products among the subjects may be due to the low availability of these food groups in low-income households. Khor *et al.* (2015) stated that the consumption of milk by low-income children was significantly lower than that of households with high incomes (Khor *et al.* 2015). Low scores for fruits, vegetables and legumes may be due to the children's low preferences and exposure towards these food groups.

For nutrient components, they obtained both low and moderate scores for fat and sodium. Low mean scores for fat indicate that the children's fat intake was high. Based on the dietary data collected, most of the children eat fried and high-fat foods during their main meal, such as fried chicken, fried fish, fried rice, fried noodles, French fries, nasi lemak, roti canai, and burger. Fast foods such as French fries and burger have poor nutritional quality and the consumption of these foods may lead to higher calorie intake and poor diet quality among children (Nemati *et al.* 2020). In addition, fish crackers, soy sauce, chicken or beef patty and instant noodles are common high sodium foods consumed by the children. High intake of these nutrients will bring adverse effects to health if not controlled.

This study analyzed socio-demographic characteristics and its association with diet quality. Only one socio-demographic variable was associated with diet quality, which is the number of children per household. The size of the family including number of children in a family determines their nutritional consumption in such a way that the allocation of food per child is likely to decrease with the increase in the number of children, which, in turn, may adversely affect the nutritional status of children (Bhattacherjee *et al.* 2016). However, this study found that more children from household with less than three children had poor diet. In this study this can also partly be explained by the diminishing effect of income on diet quality, this may be due to rural

families with more income leads to increase number of children. While on the other hand, poor family had less children. In contrast with a finding from Appelhans *et al.* (2012), they found a positive association on household income and diet quality where households with lower income reported purchasing fewer fiber, fruits and vegetables but more sugary foods, in comparison with households with higher income (Appelhans *et al.* 2012).

This study found that there was no significant association found between diet quality and growth status. The findings were in contrast with a study conducted in rural Bangladesh, where the prevalence of children who are stunted is high, and the overall diet quality is likely to be low (Rah *et al.* 2010). Due to short periods of study, a regular eating pattern and usual food intake by the children cannot be obtained, which may be a possible reason for lack of association for these variables. Thus, it cannot be proven that the children's dietary intake will affect their growth since there may be other potential factors that may interfere with the children's growth.

A few limitations worth to be noted in this research. The use of statistical analysis and reflective of the general population at risk is constrained by the small sample size and convenience sampling method. The children's diet recall was obtained from the mothers, which may interfere with the accuracy of the data because the data recall relies on the respondent's memory. This can result in under-reporting or over-reporting of the children's food intake.

CONCLUSION

This research provides baseline information on the diet quality and children's growth status in Tuba Island, Langkawi. The overall low HEI score was found which indicates poor diet was prevalent in the study area. Their intake was largely made up of highly energy-dense foods with poor nutritional values. Number of children per household was associated with diet quality where family with less children had poorer diet quality. No significant association was found between diet quality and growth status. It is recommended that further research need to be carried out to determine the relationship between diet quality of these children with their growth status. Findings of this study may also benefit relevant agencies to implement education program in order to provide the right knowledge on nutrition and health for this targeted population to tackle their poor dietary habits and improve their overall well-being.

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DECLARATION OF INTERESTS

The authors have no conflict of interest.

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Physical Activity, Food Consumption, and Breakfast among Normal and Overweight Elementary School Children in Bogor during Covid-19 Pandemic

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ABSTRACT

The objective of this study was to analyze differences in physical activity, quality of food consumption and breakfast between elementary school children with normal and overweight nutritional status in Bogor City of Indonesia during the Covid-19 pandemic. This was a cross sectional study carried out from September 2020 to January 2021 in nine elementary schools in Bogor City. This research was conducted when school from home had been running for about six months. Survey was conducted using a structured questionnaire filled by the subject's parents via google form and Microsoft word and then interviewing via whatsapp. Physical activity measured using the Physical Activity Level (PAL) method and food consumption quality using the Individual Dietary Diversity Score (IDDS). Breakfast quality based on the intake and contribution of energy and protein at breakfast. Result showed that 70% of male and 30% of female subjects were classified as overweight. There was a significant difference in the PAL value between subjects with normal weight and overweight (2.02 vs 1.63, $p<0.05$). There were no significant differences in IDDS of normal weight and overweight students in both weekday and weekend ($p>0.05$). However, IDDS scores of students with normal nutritional status was higher (7.08 and 8.60) compared to (6.80 and 6.78) in overweight students during the weekday and weekend respectively. In contrast, the energy and protein intake consumed during breakfast for overweight students was higher (617 kcal/day and 21 g/day) than students with normal nutritional status (477 kcal/day and 18.2 g/day) ($p<0.05$). This study has shown the importance to educate parents of overweight subjects to increase their children's physical activity as the students with normal nutrition status do as well as to provide breakfast with a more diverse menu.

Keywords: elementary school children, food consumption, overweight, physical activity

INTRODUCTION

Human capital is an important investment for the country growth and development this started since the early age. In addition to education, health and nutrition are also important contributors for improving the quality of future human resources. However, Indonesia is currently experiencing a triple burden of malnutrition, this encompasses the problem of under nutrition, micronutrient deficiencies and the problem of over-nutrition (overweight and obesity). Obesity is an increase fat mass either in certain parts or all parts of the body, or being overweight exceeds 20% of normal body weight (Mahan & Escott-Stump 2008).

Based on data from the Ministry of Health Republic of Indonesia (MoH RI 2018) the problem of obesity in children aged 5–12 years in West Java was still high at 18.8%, consisting of overweight 10.7% and obese 8.1%. Meanwhile, the prevalence of overweight children aged 5–12 years in Bogor City was 17.2%. Several factors that have been associated with obesity in children were sedentary lifestyle, unhealthy environment and food consumption. These factors can be seen at home, at school and in the community. The current Covid-19 pandemic accentuate this unhealthy environment. The sedentary lifestyle is characterized by decreased human movement, so that the level of physical activity and energy expenditure becomes lower. This lifestyle change was thought to be one of the triggering factors for increasing overweight and obesity (Oktaviani *et al.* 2012). Annisa (2014) shown that 78.8% of overweight students in Bogor City have light activities or sedentary lifestyle.

Indonesian usually eat several meals a day, this includes breakfast, lunch, dinner and snacking. The quality of food consume during

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the meal time can be measured qualitatively by its diversity. Dewanti (2020) wrote that diversifying one's food consumption is an effort to meet their nutritional needs, because no single food contains all the nutrients. In addition, Retraningrum and Dieny (2015) showed that low quality in food consumption and lack of physical activity affect the obesity status of children and adolescents. Among all meals, breakfast was deemed as one of the most important meal for the day. Breakfast is an eating and drinking done in the morning until 9 a.m. to meet 15–30% of daily nutritional needs. Breakfast should meet 300–500 kcal and 6–10 g of protein (Hardinsyah & Aries 2012). Milimet *et al.* (2010), stated that skipping breakfast can increase risk for weight gain by triggering eating more food during the day and at night. According to Mariza and Kusumastuti (2013) children's breakfast habits can affect children's snacking habits, where children who do not usually eat breakfast can increase the risk of snacking by 1.5 times. Students who skip breakfast tend to consume more snacks with higher calories.

Obese children have high risk of becoming obese as adults and have the potential to experience chronic non communicable diseases, including cardiovascular disease, hypertension, diabetes and others (Agustina *et al.* 2019). Based the background mentioned above, this research aimed to analyse differences in physical activity, quality of food consumption and breakfast between elementary school children with normal and overweight nutritional status in Bogor City during pandemic of Covid-19. The data was taken in Bogor City because the prevalence of overweight and obesity in children in the city were high. The context of Covid-19 pandemics adds a nuance to the data collection procedure as well as its analysis.

METHODS

Design, location, and time

This research was an analytic observational study utilizing a cross sectional design conducted from September 2020 to January 2021. The School from Home (SFH) policy has been running for six months when the data collection was started. The research was conducted online in nine elementary schools in Bogor City. The study had obtained permission and approval from the Research Ethics Commission of the Institute for Research and Community Service (LPPM) IPB University with the number: 296/IT3.KEPMSM-IPB/SK/2020.

Sampling

The subjects of this study were students (grades four and five) with the age range of 10–11 years old from nine elementary schools (*Sekolah Dasar*) (SD) in Bogor city. The calculation of the minimum number of samples was based on to the proportion children with normal nutritional status and overweight with low physical activity which was 31.8% and 68.2% respectively, Rahma and Bambang (2020). The minimum number of subjects obtained was 38 children for each group. However, to anticipate drop out, the total number of participants was increased to 50 children with normal nutritional status and 50 children with overweight nutritional status (overweight and obesity). The sampling technique in this study was convenience sampling, Etikan *et al.* (2016) stated that convenience sampling is used for both qualitative and quantitative studies although it is most often used in quantitative studies. Convenience sampling is a type of nonrandom sampling in which members of the target population selected as sample are those who meet certain practical criteria such as easy accessibility, geographic proximity, availability at a certain time or willingness to participate.

Sampling selection was done through several steps, first is to obtain permission from the school principals. The principal then mandated the homeroom teacher to convey information related to the research to parents of students via WhatsApp messages. Parents who are willing to become respondents then join a special WhatsApp group for the research. The data collection was done consecutively one school at the time. Each school gets a different number of respondents. The number of respondents in each school are as follow: SD Insan Kamil six people (four normal nutritional status and three overweight), SD Sinar Indonesia two people (one normal nutritional status and one overweight), SD Bina Bangsa Sejahtera 11 people (eight normal nutritional status and three overweight), SD Al-Mustarih 20 people (eight normal nutritional status and 12 overweight), SD Bosowa Bina Insani 19 people (13 normal nutritional status and six overweight), SD Aliya 17 people (eight normal nutritional status and nine overweight), SD Al-Munawwar

five people (three normal nutritional status and two overweight), SD IT-ABN 12 people (six normal nutritional status and six overweight), and SD Insantama eight people (0 normal nutritional status and eight overweight). The total subjects participated in this study were 100 people.

Data collection

The data collection method was adapted to the School from Home (SFH) condition. During the Covid-19 pandemic, the school and parents did not give permission for researchers to meet at home or at school. Therefore, the measurements of height and weight were carried out by parents at their respective homes. Weight was measured using home scale and height was measured using measuring tape. Subject were group based on the anthropometric screening results of weight and height to obtain their Body Mass Index (BMI). The nutritional status group was seen based on the BMI's Z-score. Subjects who had z-score value of -2SD to +1SD are included in the normal nutritional status group. Subjects who had z-score value of +1SD to +2SD and >+2SD are included in overweight nutritional status group (Permenkes RI 2020).

The data on subjects' characteristics, physical activity and food consumption were filled out by parents through a questionnaire in google form and Microsoft Word, then a follow up interview was conducted by phone. The children did not fill out any of the questionnaire because measurements and questionnaire filled by mothers were considered as more valid, since mothers were responsible for providing food for their children every day.

Data on physical activity collected includes the type of activity and the time allocation for each activity. Physical activity data consisted of the average physical activity on weekday and weekends. Physical activity was expressed in terms of physical activity level or Physical Activity Level (PAL). PAL is the amount of energy expended (kcal) per kilogram of body weight in 24 hours. PAR (Physical Activity Rate) was the amount of energy expended for a certain type of activity per unit of time. PAR values differ from one activity to another. PAL was calculated by the total multiplication formula of PAR with the time allocation for each activity then divided by 24 hours. Physical Activity Level (PAL) was categorized into four, namely: very light (PAL ≤1.39); light (PAL 1.40–1.69); moderate (PAL 1.70–1.99); and heavy (PAL 2.00–2.40) (FAO/WHO/UNU 2001).

Food consumption data was obtained through 2x24 hours food recalls on weekday and weekend which then translated into Individual Dietary Diversity Score (IDDS). The diversity of food consumption was calculated based on nine food groups, namely starchy staple foods; green vegetable; fruit and vegetable sources of vitamin A; fruit and vegetables and others; innards; meat. fish and chicken; egg; pods, beans, and seeds; and milk and its processed products. Any food group consumed for ≥10 g was assigned a score of one, while score of zero was assigned for consumption of less than 10 g. Kennedy *et al.* (2007) dietary diversity based on IDDS can be grouped into three categories, namely low diversity (≤3 types of food groups/day), moderate diversity (4–5 types of food groups/day) and high diversity (≥6 types of food groups/day). Data regarding calorie contribution from breakfast were obtained from food record in a week.

Data analysis

Data processing and analysis was carried out using Microsoft Excel 2017 and SPSS version 17.0 for Windows. Associations between the children's nutritional status with categorical variables were tested using chi-square test. Kolmogrov Smirnov test was used to check for normality. Independent sample t-test was used for data with normal distribution. While, Mann-Whitney test was used for non-normally distributed data.

RESULTS AND DISCUSSION

Characteristics of subjects

Table 1 showed that there were significant differences between the gender of normal and overweight subjects ($p<0.05$) with more male (70%) were overweight compared to female (30%). A study among elementary school children in Banda Aceh by Rahmad's (2019) showed that there were more female (59.5%) with obesity compared to male (40.5%). However, it should bear in mind that this study focused on overweight rather than obesity and used convenience sampling. As shown in Table 1, there was no significant difference in the age of subjects with normal nutritional status and

Table 1. Characteristics of subjects based on nutritional status

Characteristics of subjects	Normal (n=50)	Overweight (n=50)	p
	n (%)		
Sex			
Male	26 (52.0)	35 (70.0)	0.033*
Female	24 (48.0)	15 (30.0)	
Age (years old)			
10	36 (72.0)	35 (70.0)	
11	12 (24.0)	15 (30.0)	0.155
12	2 (4.0)	0 (0.0)	
Allowance (IDR/day)			
<IDR10,000	1 (2.0)	3 (6.0)	
IDR10,000–19,999	46 (92.0)	45 (90.0)	0.273
≥IDR20,000	3 (6.0)	2 (4.0)	

Chi-square test; *Significance p<0.05
IDR: Indonesian Rupiah

overweight nutritional status (p>0.05). Most of the subjects were ten years old, both in normal subjects (72.0%) and overweight subjects (70.0%). According to Ermona and Wirjatmadi (2018), children at the age of ten both boys and girls, have an appetite that tends to increase during their growth period towards adolescence. The results showed that there was no significant difference between the daily allowances of normal and overweight subjects (p>0.05). The majority of the normal subjects (92.0%) and overweight subjects (90.0%) were given pocket money of IDR10,000–IDR19,999 per day. According to Faghih *et al.* (2015) children who get a larger amount of pocket money tend to consume fast food more often which lead to unhealthy lifestyle as one of the factors causing obesity.

Physical activity

Physical activity was body movement characterized by the work of skeletal muscles and increased energy and energy expenditure (MoH RI 2018). Table 2 showed the average of activity duration between normal and overweight subjects on weekday and weekend during the Covid-19 pandemic.

Table 2 showed that there were significant differences in duration of sleep, personal activity, online learning, playing and light activity between normal and overweight subjects (p<0.05). Overweight subjects had significantly longer duration of sleep, online learning and light activities (except for playing Lego) compared with normal subject. While, normal subject had significantly longer duration of personal activity and playing than overweight subjects. Na'imah's research (2014) showed that subjects who got enough sleep (7–9 hours a day) tended to have a normal body mass index compared to subjects who have less sleep duration (<7 hours a day).

Spaeth *et al.* (2019) stated that less sleep is associated with higher Body Mass Index (BMI). Sleep problems can affect the body's hormones and metabolism. In addition, Muscogiuri *et al.* (2019) found that increased intake of unhealthy food consumption occurs when hormones and metabolism were in abnormal conditions and it can trigger weight gain.

The average duration of watching TV and playing gadgets was significantly longer in overweight subjects than normal subject. The average screen time for both groups during the Covid-19 pandemic was more than two hours per day. In contrast, Xiang *et al.* (2020) found that before Covid-19 pandemic about 92.7% of children and adolescents in China had screen time duration of two hours per day or less while only 7.3% had screen time duration of more than two hours per day. American Academy of Pediatrics (AAP) (2001) stated that Low Screen Time (LST) is a screen time with a duration of <2 hours per day and High Screen Time (HST) with a duration of two hours per day. Chassiakos *et al.* (2016) also recommends screen time duration for children and adolescents as much as two hours per day.

Screen time viewing was one of the external factors that influence diet trough exposure to and promotion of unhealthy diets and lifestyle. Tarabashkina *et al.* (2016) stated that screen time can cause higher energy intake, one of which was caused by exposure to food and beverage advertisements that are widely offered on television. The products offered in advertisements are generally foods that contain high energy. Children exposed to advertisements tend to consume the advertised food so that they have a higher energy intake than children who

Table 2. The differences of activity duration between normal and overweight subjects

Activities	Normal (hours)	Overweight (hours)	p
Sleep	8.85±0.18	9.71±0.50	0.000[1*]
Personal	2.89±0.11	2.66±0.13	0.000[1*]
Praying	1.64±0.11	1.40±0.13	0.000[1*]
Take a bath	0.50±0.00	0.51±0.03	0.047[1*]
Eat	0.75±0.00	0.75±0.00	0.500[1]
Online learning	2.64±0.31	3.34±0.60	0.000[1*]
Carry out task	0.89±0.31	1.59±0.60	0.000[1*]
School from home	1.75±0.00	1.75±0.00	0.500[1]
Playing	5.00±0.67	1.89±0.60	0.000[1*]
Cycling	2.09±0.47	0.93±0.50	0.000[2*]
Badminton	0.43±0.35	0.03±0.12	0.000[1*]
Football	0.51±0.59	0.15±0.25	0.001[1*]
Ping pong	0.57±0.50	0.27±0.31	0.001[1*]
Jumping rope	0.21±0.38	0.02±0.07	0.001[1*]
Hide and seek	1.19±0.56	0.49±0.33	0.000[1*]
Light activity	4.62±0.53	6.40±0.78	0.000[2*]
Watching television	2.16±0.50	3.53±0.75	0.000[2*]
Playing gadget	1.26±0.42	2.15±0.56	0.000[2*]
Playing lego	1.20±0.76	0.72±10.76	0.001[2*]

[1]Mann-whitney test; [2]Independent sample t-test; [*]Significance p<0.05

were not exposed to advertisements. Children and adolescents consumed foods which was high in sugar, salt, fat, calories and carbonated drinks (Wardlaw & Hampl 2007). According to Pinho *et al.* (2017) poor diet can increase BMI and causing obesity. In addition, longer screen time mean less time for physical activity and sedentary behavior in adolescents was one of the risk factors that can cause obesity. Further, Laurson *et al.* (2014) stated that screen time can cause sleep disturbances. This was caused by the presence of artificial light from the screen so that it interferes with the hormonal response. Decreased sleep time, one of which is caused by artificial light from excessive screen time, thereby increasing sleep deprivation and increasing energy intake. According to Bel *et al.* (2013) adolescents who sleep less than eight hours have higher fat intake, consumption of foods high in energy content and low quality of diet (foods with high energy density and low nutrient content) when compared to adolescents who sleep ≥8 hours a day. During pandemic of Covid-19, subject distribution based on their physical activity level can be seen in Table 3.

Table 3 shows that there was significant difference in physical activity between normal and overweight subjects (p=0.000). During the Covid-19 pandemic, 64.0% of normal subjects had heavy physical activity with PAL value of 2.02. While on the other hand, most of the overweight subjects (90.0%) had light physical activity with PAL value of 1.63. The results showed that during pandemic of Covid-19, normal subject had better physical activity than overweight subject. Similarly, Rizkiyah (2015) showed that normal subjects had higher PAL value than overweight subject, despite both groups were classified as having light physical activity. In addition, Jiménez-Pavón (2010), also found that children with low physical activity levels had higher Body Mass Index than normal and had greater chance of experiencing more nutritional problems. Colley *et al.* (2013),

Table 3. Subject distribution based on physical activity

Physical activity level (PAL)	Normal		Overweight		Total	
	n	%	n	%	n	%
Very low (≤1.39)	0	0.0	0	0.0	0	0.0
Low (1.40–1.69)	0	0.0	45	90.0	45	45.0
Moderate (1.70–1.99)	18	36.0	5	10.0	24	24.0
Severe (2.00–2.40)	32	64.0	0	0.0	31	31.0

showed that physical activity was associated with the incidence of over-nutrition in children. This happens because of an imbalance between energy intake and energy released from the body.

Quality of food consumption

Table 4 showed the difference in the average score of each food groups consumed based on the IDDS for normal and overweight subjects on school days (weekday). While Table 5 showed the same information for weekend. Children from the normal weight group had a significantly higher average scores in consumption of Vitamin A source fruit and vegetable and other fruits and vegetables ($p<0.05$). The average score of fruit and vegetable sources of vitamin A was 0.92 food groups/day in children with normal weight and 0.80 food groups/day in the overweight group. Normal subject had an average score of 0.98 food groups/day for other fruits and vegetables while the overweight group was 0.84 food groups/day. This implies that, more students in the normal weight group consumed vitamin A source fruits and vegetables as well as other fruits and vegetables. However, there was no difference in the total score of IDDS between normal and overweight subjects ($p=0.227$) and both groups had a high IDDS based on Kennedy *et al.* (2007) since both groups consumed more than 6 types of food groups/day. The average IDDS for children in the normal weight group was 7.08 (SD 1.96) while in the overweight group was 6.80 (SD 2.75).

In contrast to the IDDS during the weekday, on the weekend the consumption of fruits and vegetables and vitamin A source food showed no significant difference in both groups. The total IDDS also showed no significant difference between the two groups. The IDDS average score in the group with normal weight was 8.60 (SD 5.95) food groups/day or higher compared to the weekday, and among the overweight it was 6.78 (SD 2.73) food groups/day or almost the same as the weekday. This showed that during the weekend, children with normal weight tend to eat more food groups.

Table 4. The difference of average score for diversity of food consumption in normal and overweight subjects based on food groups during weekday

Food group	Individual dietary diversity score		p
	Normal (Mean±SD)	Overweight (Mean±SD)	
Starchy staple food	1.00±0.00	1.00±0.00	0.500
Green vegetable	0.90±0.30	0.90±0.30	0.500
Fruits and vegetables sources of vitamin A	0.92±0.27	0.80±0.40	0.043*
Other fruits and vegetables	0.98±0.14	0.84±0.37	0.008*
Innards	0.00±0.00	0.04±0.20	0.078
Meat, fish, and poultry	1.00±0.00	0.96±0.20	0.078
Egg	0.62±0.49	0.66±0.48	0.266
Pod, peanuts, grains	0.80±0.40	0.74±0.44	0.239
Milk and processed products	0.86±0.35	0.86±0.35	0.500
Individual dietary diversity score (IDDS)	7.08±1.96	6.80±2.75	0.227

Mann-whitney test; *Significance $p<0.05$

Table 5. The difference of dietary diversity score between normal and overweight subjects based on food groups during weekend

Food group	Individual dietary diversity score (IDDS)		p
	Normal (Mean±SD)	Overweight (Mean±SD)	
Starchy staple food	1.00±0.00	1.00±0.00	0.500
Green vegetable	0.92±0.27	0.90±0.30	0.364
Fruits and vegetables sources of vitamin A	0.90±0.30	0.90±0.30	0.500
Other fruits and vegetables	0.80±0.40	0.86±0.35	0.214
Innards	0.06±0.24	0.08±0.27	0.349
Meat, fish, and poultry	1.00±0.00	0.98±0.14	0.159
Egg	0.54±0.50	0.56±0.50	0.421
Pod, peanuts, grains	0.86±0.35	0.82±0.39	0.294
Milk and processed products milk	0.70±0.46	0.68±0.47	0.415
Individual dietary diversity score (IDDS)	8.60±5.95	6.78±2.73	0.468

Mann-whitney test; *Significance p<0.05

The study of Swamilaksita and Sa'pang (2018) showed that there was no significant difference in IDDS in children with normal nutritional status and obesity (p=0.791). However, in their study the mean IDDS was very low at only 1.5 (SD 0.5) food groups/day. The food groups most consumed by school children in West Jakarta in the study are sources of carbohydrates (bread, rice, potatoes), sources of animal protein (sausage, eggs, chicken), and sources of vegetable protein (*tempe*, green beans, tofu). Similarly, Nurrachmat (2016) also found no significant difference in the diversity of food consumption between normal and obese school children (p=0.705) with the average IDDS of five food groups/day in children with normal weight and 4.89 food groups/day in children with obesity. The most common food groups consumed in the study were starchy staple foods; meat, fish, chicken; and eggs.

Breakfast quality

Table 6 showed the intake and contribution of energy and protein from breakfast for normal weight and overweight subjects. The energy intake from breakfast for overweight subjects (617 kcal) was greater than normal subjects (477 kcal). Protein intake for overweight subjects (21 g) was higher than normal subjects (18.2 g). According to Hardinsyah (2012) breakfast fulfills about 15–25% of daily nutritional needs. Breakfast energy intake should meet 300–500

Table 6. Differences energy and protein intake from breakfast between normal and overweight subjects

Energy and Protein	Normal	Overweight	p[1]
	Mean±SD		
Energy			
Intake (kcal/day)	477±71.7	617±140.1	0.000*
Contribution to daily intake (%)	28.4±3.3	27.6±4.1	0.157
Contribution to energy adequacy (%)	29.9±6.3	31.3±6.9	0.153
Protein			
Intake (g/day)	18.2±2.8	21.0±3.9	0.000*
Contribution to daily intake (%)	30.5±6.6	28.6±5.8	0.067
Contribution to protein adequacy (%)	41.0±8.4	42.4±8.3	0.199

[1]Independent sampel t-test; *Significance p<0.05

kcal/day and protein intake 6–10 g/day. Thus, results showed that energy intake from breakfast for children with normal weight was within the recommended range, while for the overweight subjects it exceeded the recommended range. On the other hand, the protein intake from breakfast in children in both groups exceeded the recommended limit.

Table 6 showed that during pandemic of Covid-19, there was no difference in the contribution of energy to daily intake and energy adequacy in the two groups of subjects ($p>0.05$). Likewise, there was no difference in the contribution of protein to daily intake and protein adequacy in the two groups of subjects ($p>0.05$). In normal subjects, breakfast contributed energy and protein to the daily intake greater than the overweight subjects. In normal subjects, the contribution of breakfast contributed energy and protein to nutritional adequacy was greater than the overweight subjects. The results of the study were in accordance with Ifdal (2014), who found that among university students the energy intake from breakfast in overweigh subject (425 kcal/day) was greater than normal subject (365 kcal/day). The protein intake from breakfast of overweight subject (10.1 g/day) was greater than normal subject (9.6 g/day). A study on school children in Banda Aceh showed that 59.5% of obese school children had a poor breakfast intake. Breakfast intake is considered inadequate if it contributes less than 200–300 kcal/day or more than 200–300 kcal/day. In the study, more than half (57.1%) of school children with normal nutritional status had adequate breakfast intake of around 200–300 kcal/day (Rahmad 2019). Our study found that the intake of energy and protein was higher among our subjects during the pandemic compared to previous study, this might be because children are more likely to consume staple food, meat, fish and poultry than fruits and vegetables during this time.

The was conducted during the Covid-19 pandemic, which has not been widely carried out. Process of calculating the difference in the duration of each activity carried out by the subject as well as the quality of food consumption using the IDDS method. This can indirectly educate parents and increase their awareness on the importance of physical activity and dietary diversity for their children. However, due to the mobility restriction during the Covid-19 pandemic, the anthropometry measurement of the subject's weight and height was not carried out by the researcher and the subject population only came from private schools due to the use of convenience sampling method.

CONCLUSION

In this study found more male subjects with overweight status than female. During pandemic of Covid-19, subjects with normal nutritional status had higher level of physical activity (PAL 2.02) than subjects with overweight nutritional status (PAL 1.63). Overall, the total score of diversity from normal subject was higher than of overweight subject (weekday=7.08; weekend=8.60). The intake of energy and protein from breakfast was significantly higher in overweight subject (617 kcal energy and protein 21 g/day) than in normal subject (477 kcal energy and 18.2 g protein/day). Recommendations for further research, to improve sampling method to cover more diverse demography, to get more valid results it is better to measure height and weight directly by researchers and measure breakfast quality based on macronutrients (energy, protein, fat, and carbohydrates).

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DECLARATION OF INTERESTS

The authors have no conflict of interest.

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Facilitators and Barriers to Breastfeeding Practices of Mothers in a Selected Urban Community in Malaysia

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ABSTRACT

This study was designed to explore the facilitators and barriers of breastfeeding practices among mothers in a selected urban population in Malaysia. This study reports the qualitative findings from a larger study that was conducted in two health clinics in Selangor, Malaysia. Ten mothers were recruited and an in-depth interview using grounded theory approach was used to obtain information. Interviews were transcribed and was manually arranged into codes and themes using thematic analysis. Facilitators to breastfeeding in this population were maternal factors (knowledge of breastfeeding, intention to breastfeed, perceived benefits of breastfeeding), social support and health care provider. Barriers to breastfeeding were insufficiency of breastmilk, employment and stigma of public breastfeeding. The findings of this study suggest that intervention to improve breastfeeding uptake should aim on increasing knowledge of mothers on coping with issues related to breastfeeding difficulties such as how to increase or sustain milk production and increasing awareness of employers and public on supporting breastfeeding mothers.

Keywords: breastfeeding, early life nutrition, infant nutrition, maternal experience, qualitative

INTRODUCTION

Breastfeeding supports healthy physical and cognitive development of a child (Wallenborn *et al.* 2021). Breastfeeding is also associated with development of food preferences, hunger regulation and promotes healthy mental health in a child (Verduci *et al.* 2014; Adan *et al.* 2019; Nguyen *et al.* 2021). Breastmilk contains bioactive compounds such as hormones, cytokines, immunoglobulins and other beneficials compounds that increases defence against harmful pathogens and increases tolerance towards antigens (Carr *et al.* 2021). Recent studies documented that breastfeeding protects against future obesity and non-communicable diseases such as hypertension, diabetes and atherosclerosis (Verduci *et al.* 2014; Binns 2016). Studies also show that breastfeeding helps to reduce the risk of ovarian cancer, breast cancer and important in spacing out child (Victoria *et al.* 2016).

The World Health Organization (WHO) has recommended that infants should be exclusively breastfed for the first six months and followed by introduction to solids from six months on words with the continuation of breastfeeding untill two years of age (WHO 2020). However, adhering to the recommendations still remains a challenge (United Nations Children's Fund (UNICEF) 2019).

Global report indicated that only 41% of infants under six months old are exclusively breastfed, 71% were breastfed at one year of age and only 45% were breastfed at two years of age worldwide (UNICEF/WHO 2017). In Malaysia, the National health and Morbidity Survey in 2016 reported that only 47.1% of the infants in this country were exclusively breastfed until six months while only 39.4% of the infants were breastfed untill two years old (National Health and Morbidity Survey 2016).

A mother's successful breastfeeding journey is determined by factors involving herself, her child and people around the mother. Maternal education, age, perception, experience, nature of birth and external factors such as family support, involvement of husband and grandmothers, peers and etc., play an important role in the success of breastfeeding among mothers (Emmott &

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Mace 2015; Martin *et al.* 2020; Theodorah & Mc'Deline 2021). Studies show that mothers that receive support from their mothers, mothers in law, husband and friends tend to breastfeed longer compared to those who did not receive these support (Martin *et al.* 2020; Theodorah & Mc'Deline 2021).

The external environment such as the involvement of healthcare practitioners, policies revolving breastfeeding and maternity leave, breastfeeding facilities in working premise and shopping mall play an important role in sustaining breastfeeding in mothers (Rollins & Doherty, 2019). Studies show inconsistent findings on the level of education and support received from healthcare practitioners worldwide (Chang *et al.* 2020; (Fox *et al.* 2015). Some studies have reported an increase in breastfeeding prevalence after the constant education done during prenatal appointment while there are studies that have reported a poor initiation of breastfeeding even after childbirth despite having received prenatal education on breastfeeding (Mehlawat *et al.* 2020). Limited maternity leave and absence of breastfeeding facilities in workplace also remains a barrier to breastfeeding (Febrianingtyas *et al.* 2019).

A large pool of literature exists on the facilitators and barriers of breastfeeding globally. However, the way of how these factors influence mothers throughout breastfeeding differs according to population, country, degree of urbanization, religion and etc. Hence identifying which and how all these factors influence a mother's experience in breastfeeding in a population will help in planning effective interventions to encourage breastfeeding in the targeted population.

Hence, this study was designed to explore the predictors and barriers of breastfeeding practices among mothers in a selected urban population in Malaysia. The qualitative design of the study was able to identify issues that had been overlooked in a multi ethnic setting like Malaysia. The multi ethnic Asian platform was able to highlight cultural norms and obstacles faced by these mothers. The results, of this study may serve a guideline to understand the predictors and barriers of breastfeeding in this country and other multi ethnic population globally to improve existing policies and guidelines to improve prevalence of breastfeeding.

METHODS

Design, location, and time

The data from this study was obtained from a larger study that was conducted in 2019. The original study aimed to assess feeding practices and growth among the infants. It was a mixed methods study involving quantitative and qualitative design. In the quantitative part of the study, prospective data on infant feeding and growth were obtained while in the qualitative part, issues regarding infant feeding were obtained through in-depth interviews using a grounded theory approach. This study reports the qualitative findings from the bigger study.

The study was conducted in two community health clinics in Klang Valley Selangor. The two community health clinics were in Meru and Puncak Alam. The location was decided based on the availability of high number of mother infants' pairs from all the three ethnic groups that has been decided as the inclusion criteria of this study. One hundred fifty mother infant pairs were recruited in the larger study, in which ten mothers were enrolled for the qualitative part. This study reports the findings from the in-depth interviews.

A grounded theory approach was used to identify, analyse and develop a theoretical framework that describes predictors and barriers in breastfeeding practices among the study population (Glaser & Strauss 2009). Mothers of the infants who agreed to join the qualitative part of the study were interviewed using an in-depth interview method. A semi structured interview guide was used to facilitate the interview to ensure information regarding the areas of interest were obtained.

Ethical approval for the original study was obtained from the National Medical Research Register (NMRR), reference No: KKM/NIHSEC/PIT-811, prior to commencing any field work. Informed consent was read in either English or Malay, before obtaining participant's consent through a signature.

Sampling

All the ten participants included in this study were mothers of infants aged six to eleven months old. All their infants had a birth weight of 2,500 g to 4,000 g and were full term infants (37 to 42 gestation weeks) from singleton pregnancy. They were selected based on their willingness to

participate in the in-depth interviews and also based on the emerging themes such as employed and unemployed mothers, mothers with lactation issues, mothers who consulted healthcare professionals and other immediate people to seek information and etc throughout data collection. Mothers whose infants were admitted for three days or more, two weeks before data collection or diagnosed with any congenital, medical or digestive problems that may impair their dietary intake were excluded.

Participants were given pseudonyms according to breastfeeding status at the point of data collection (NB for non-breastfeeding, and B for breastfeeding) and their ethnic group (M for Malay mothers, I for Indian mothers and C for Chinese mothers) followed by hierarchy of point of contact (one for first mother interviewed, two for second mother interviewed etc).

Data collection

Participants whom the primary investigator or Co researcher identified as suitable to be recruited in the qualitative part of the study based on the emerging themes were approached. Participants were interviewed by either the Primary investigator or Co-researcher who were females, only if they agreed to be involved in the qualitative part of the study. These participants were interviewed in either English or Bahasa Melayu (national language) using an in-depth interview method guided by a semi structured interview guide.

The semi structured interview guide was designed to enable us to capture the various facilitators and barriers to breastfeeding. The questions were designed to explore conditions that may affect breastfeeding practices that may promote or obstruct mothers from breastfeeding. These questions were initially informed by the literature and included: (a) Duration of breastfeeding and non-breastfeeding practices; (b) Knowledge seeking on breastfeeding; (c) Reasons for quitting or sustaining breastfeeding; (d) Support received on breastfeeding. Along the way, probes and extra questions were added guided by a grounded theory approach.

Participants were approached and interviewed till data saturation was reached, in which there were no newer codes or themes that emerged from the interviews. Each in depth interview lasted for 30 minutes to an hour. Observations were recorded as field notes in a journal throughout the interviews.

Data analysis

The audio interviews were transcribed verbatim. Interviews were translated to English language and sections from the transcripts that were unclear were checked back with the audio recording. Data was sorted and coded manually. The interviews were first coded by the Co researcher and later was independently read and coded by the principal investigator. Codes were arranged into themes guided by a thematic analysis approach (Braun & Clarke 2006).

RESULTS AND DISCUSSION

Maternal sociodemographic and prenatal characteristics

A total of ten mother infant pairs were involved in this study. Most of the mothers were between 30 to 34 years old. Mothers were largely from the Malay ethnic group (n=7) while the remaining (n=3) were from the non-Malay ethnic group. Most mothers, (n=7) had a secondary school education while only one of them had a primary school education while the remaining two mothers had a tertiary school education. Majority of the participants, (n=7) reported a household monthly income of more than USD719 (RM3,000) while only one participant reported an income of less than USD239.5 (RM1,000). The study also indicated that Most mothers (n=6) worked after delivery. The participants' characteristics are shown in Table 1.

Most mothers in this study (n=6), had a normal pre-pregnancy Body Mass Index (BMI), (18.5–24.9 kg/m^2). The study indicated that (n=5) of the mothers fell 'under recommended weight gain' group, (n=3) of the mothers fell within a 'recommended weight gain" category, while (n=2) of the participants fell in the "over the recommended weight gain" group. Maternal prenatal factors are included in Table 1.

Infant characteristics

There were six male infants and four female infants involved in this study. Most infants were not the first child (n=7) and mothers reported that all of them had breastfed their child. However only (n=6) of the infants were exclusively breastfed till six months. Three of them were

Table 1. Maternal sociodemographic and prenatal characteristics

Variables	Frequency (n)
Age (years)	
25–29	2
30–34	6
35 and above	2
(32.10±3.178)	
Ethnicity	
Malay	7
Non-Malay	3
Education level	
Primary school	1
Secondary school	7
Tertiary level	2
Household income	
<239.5USD (RM1,000)	1
239.5–718.5USD (RM1,000–RM2,999)	2
≥719USD (RM3,000)	7
Work after delivery	
Yes	6
No	4
Pre-pregnancy BMI (kg/m^2)	
18.5–24.9	6
25.0–29.9	3
≥30.0	1
(24.9±3.687)	
Gestational weight gain	
Under recommended weight gain	5
Within recommended weight gain	3
Over the recommended weight gain	2
(6.03±6.65)	

RM; Ringgit Malaysia; USD: United States Dollar; BMI: Body Mass Index

introduced to solids before six months of age. Infants' characteristics are shown in Table 2.

Facilitators and bbarriers to breastfeeding among mothers in this study

The results on the main themes and subthemes that emerged on the predictors and facilitators of breastfeeding are given in Figure 1. Three major themes emerged on the area of predictors for breastfeeding among the participants: maternal factors, social support and healthcare provider while the themes that emerged on the area of barriers of breastfeeding perceived were lactation problem, employment, and stigma of public breastfeeding. The model developed from the themes and codes obtained from this study is shown in Figure 1.

Facilitators to breastfeeding

Maternal factors

Knowledge of breastfeeding. Knowledge on breastfeeding emerged as a subtheme for predictors of breastfeeding. Mothers in this study were aware of the importance of breastfeeding to their infants. A common importance of breastfeeding that they were aware of was the importance of breastfeeding to protect the child from common illness such as flu, fever and other diseases. This knowledge, was a motivator for some of the mothers to breastfeed their infants.

"*I breastfeed my baby until six months. I planned to breastfeed my baby because I know breastmilk is very nutritious and could protect my baby from getting a fever, flu, and other diseases*". BFM1.

Intention to breastfeed. An important predictor for breastfeeding in this study was having an intention to breastfed from pregnancy. The interviews suggested that mothers who had exclusively breastfeed their infants until 6 months and plan to continue breastfeeding until two years of age, claimed to have planned this since they were pregnant. This was apparent in the conversation of two mothers:

"*Yes, I planned to breastfeed my baby since I was pregnant. I planned to continue to breastfeed until two years". I'm able to give my breastmilk to my baby up to eight months now and I know breastmilk is much better for my baby's growth*". BFM3.

This study found that maternal factors such as perceived benefits, knowledge and

Table 2. Infant's characteristics

Variables	Frequency (n=10) Mean±SD
Gender	
Boy	6
Girl	4
Hierarchy in siblings	
First child	3
Second and above	7
Ever breastfed (0−24 months)	
Yes	10
No	-
Continued breastfeeding (after 6 months)	
Yes	6
No	4
Exclusively breastfeeding (0−6 months)	
No	3
Yes	6
Mixed feeding	1
Introduction to formula feeding (n=9)	
Yes	6
No	3
Age introduction to formula feeding (0−24 months) (n=9)	6.6±5.0
Never	3
<6 months	3
At 6 months and above	3
Age Introduction to solid foods (at 6 months)	6.9±0.74
<6 months	1
6 months and above	9

intention to breastfeed are important predictors to breastfeeding. A study conducted among breastfeeding mothers in Kelantan, Malaysia reported that those with prior experience in breastfeeding and high scores in knowledge of breastfeeding had a higher odds of exclusively breastfeeding their infant at one week postpartum (Che'Muda *et al.* 2018). A meta-analysis also stated that maternal knowledge is associated with breastfeeding at one year postpartum (Zakarija-Grković *et al.* 2016). However, maternal intention to breastfeed has been shown to be a strong predictor to breastfeed regardless of knowledge or experience suggesting that mothers need to be given knowledge of breastfeeding during pregnancy to encourage them to have the intention to sustain breastfeeding for two years (Raissian & Su 2018).

Perceived benefits of breastfeeding. One of the facilitators of breastfeeding in this study was the perceived benefits of breastfeeding by mothers. In this study, mothers were aware that breastmilk is nutritious and helps to increase the immunity of the infants. This was apparent in the conversation of a mother who claimed that breastmilk was a nutritious food and helped to protect her infant from illness:

"*In my opinion, babies who consumed breastmilk were healthier and rarely get flu, fever and other diseases*". BFM1.

Mothers also revealed that breastfeeding was easier and more convenient than formula feeding. Breastmilk was also believed to be healthier than formula milk that was claimed to contain more sugar. In addition, mothers who participated in this study also associated benefits of breastfeeding with an advantage of reducing household expenses. Participants claimed that breastfeeding was cost effective as breastmilk was free while formula milks were expensive. This was evident in the conversation with a mother:

"*Breastfeeding can save money as formula milk is very expensive, and formula milk contained more sugar compared to breast milk*". BFM7.

Mothers in this study perceived breastmilk as a healthier choice compared to formula milk as it does not contain high sugar and helps to increase the immunity of their child. They also state that breastfeeding is easier, convenient and cost effective. These factors were associated with benefits of breastfeeding hence a facilitator to breastfeed among the mothers.

Social support

Social support plays an important role as a predictor to breastfeeding in this study. Husbands, family members, and friends were important factors that influence mothers to successfully

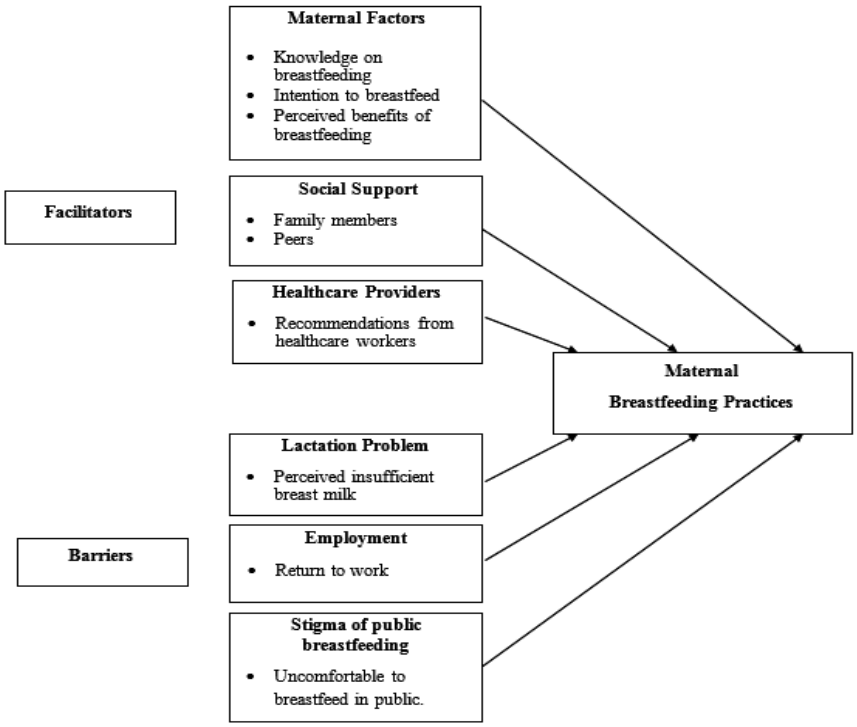

Figure 1. Model for facilitators and barriers to maternal breastfeeding practices in a selected urban community in Malaysia.

breastfeed. Most participants claimed that they learned about breastfeeding from their family members, especially their mothers and also friends. Conversation with an Indian mother revealed the encouragement given by her mother to continue breastfeeding by associating the importance of breast milk as a "gift from god".

"*My mom encouraged me to continue to breastfeed my baby, and, when I stopped breastfeeding, my mother scolded me as she said that breastmilk is a gift from God*". BFI3.

Peer support too emerged as an essential factor that played a role as a predictor for breastfeeding. Exchanging problems and solutions among friends who are also breastfeeding was important in the breastfeeding journey. This was narrated by one of the mothers:

"*Oh, I learned a lot about breastfeeding from my friends. It is much convenient to ask my friends because most of my friends have a baby around the same age as mine. We always ask for each other's opinion*". BFM4.

The role of a husband is very important in helping a mum to sustain her breastfeeding journey. Most mothers in this study expressed that they received good support from their husband to breastfeed. One of the mother, UM1 explained that her husband use to massage her back after she breastfeeds her baby. This finding further highlights that husbands were very supportive in encouraging their wives to breastfeed.

In this study, the immediate people around a mother, such as grandmothers, husband and friends were important factor for a successful breastfeeding journey among the participants. Mothers need to be comfortable, and have a support system in which they can communicate their problems and ask for suggestions from other. Having someone who is approachable and have experience in the area of breastfeeding kept the mothers motivated to continue their breastfeeding practices.

Studies show that support from family members, relatives, husband and peers may determine the success or failure of breastfeeding among the mothers (Martin *et al.* 2020). This depends on how supportive or discouraging this inner circle of people are to the mothers. Although many of them are aware of the importance of breastfeeding and do advice on how to cope with an issue during breastfeeding, studies show that there are some of them who would encourage

formula feeding or to introduce complementary feeding before six months (Theodorah & Mc'Deline 2021).

Husband or father of the child can be a barrier or support to breastfeeding. In developing countries, presence of husband is associated with early termination of breastfeeding while in developed countries a mother may not be successful in breastfeeding her child for a period of six months without the support of her husband (Emmott & Mace 2015). This shows that the perception on breastfeeding varies according to population or location.

Healthcare provider

Healthcare providers, such as doctors and nurses, played an important role as predictors to breastfeeding among the mothers in this study. All the mothers involved in this interview narrated that they have been educated on the importance of breastfeeding. They were also taught on how to breastfeed during their pregnancy. Although the interviews reported the involvement of healthcare team in educating the mothers on breastfeeding, but the role of nurses were highlighted in most of the conversation with the mothers. Interview revealed that mothers were taught on breastfeeding in a seminar during their pregnancy:

"*I learned about breastfeeding from my family members and nurses, and I did attend the seminar on breastfeeding. The nurses taught us how to breastfeed in that seminar*". BI1.

In certain cases, the nurses were very encouraging on breastfeeding. One mother revealed that she was encouraged by one of the nurses to breastfed her child within the first hour, just after her childbirth:

"*On the first day of my delivery, I had started to breastfeed him because the hospital staff encouraged me to start breastfeed within the first hour of my baby's life*". BFI3.

The important role played by the healthcare providers either at the community health clinics or at the hospitals, just after childbirth is important as a reminder and precursor for breastfeeding for these mothers to breastfeed. A study on women's experience in breastfeeding in the United Kingdom reported a mixed response towards the involvement on healthcare providers in their breastfeeding experience. Mothers complained that healthcare providers did not elaborate on the challenges and difficulty of breastfeeding resulting in mothers were unprepared and stressed after their delivery. The prolonged challenges and frustration of not being able to meet the demand of their breastfeeding child resulted in them to quit breastfeeding (Fox *et al.* 2015).

Barriers to breastfeeding

Lactation problem

Perceived insufficient breastmilk was a common barrier for mothers to exclusively breastfeed for the first six months or to continue breastfeeding until two years. They were four women in this study that claimed they stopped breastfeeding because they did not have sufficient breastmilk.

"*I don't breastfeed my baby anymore… the longest period I breastfeed my previous baby was up to 10 months, but for this baby, I have stopped at four months… I realized that I just don't have breastmilk and it's just too little*.", stated another mother. NBFM6.

The findings also revealed that, mothers did try to express their breastmilk to breastmilk their infant, however this method also was not successful hence resulting them to switch to formula milks.

"*I had to stop breastfeeding my child when he was eight months. Because I realize that my milk was insufficient. And if I pump, I was only able to get one oz only*", said one mother. NBFM5.

Perceived insufficient breastmilk is a common problem linked with early caseation of breastfeeding. In this study, this factor emerged as a barrier to breastfeeding. However, the point that mothers did try to express their breastmilk via a pump suggest that they tried their best to provide breastmilk to their child, but the fact that they still could not yield enough milk made them to stop breastfeeding.

Perceived insufficient breastmilk had always been a reason for early termination of breastfeeding. A recent study in Taiwan , involving 1,000 participants reported that nearly 40% of the mothers stopped exclusively breastfeeding at one month postpartum as they thought they could not produce sufficient milk (Chang *et al.* 2019). Previous study also reported a consistent findings in which perceived insufficient breastmilk was among the top three reason for early cessation of breastfeeding (Wagner *et al.* 2013). Addressing

maternal concern on insufficient breastmilk may help to reduce the tendency to stop breastfeeding early in life.

Employment

Employment emerged as an important barrier to breastfeed among the participants. In this study, Mothers claimed they did not have enough time to express their breastmilk to be given to their infants during working hours. In addition, working resulted in them being tired hence they were not able to get up in the night to express their breastmilk, thus resulting them to stop breastfeeding and introduce formula milk to their infants.

"*I stop breastfeeding her after six months. I don't have enough time to express my breastmilk for her when I started to work, as I feel tired*". NBF12.

Mothers also claimed that their babies refused to be breastfed directly after they were given breastmilk via bottle. This was another reason, for mothers to stop breastfeeding as they claimed they were too tired to pump their breastmilk at night as they were tired after work. This slowly resulted in them to introduce formula milk to their infants as they found it more convenient. In addition, there were mothers who complained that their breastmilk production reduced drastically after they started working, resulting them to stop giving breastmilk.

Breastfeeding mothers often quit breastfeeding after resuming work, this finding has been consistent in many studies. However, increasing the length of paid maternity leave does promote breastfeeding in low- and middle-income countries (Chai *et al.* 2018). Employed mothers also reported feeling tired and sleep deprived and the challenge of finding time to pump milk during working hours too resulted in poor milk production which eventually leads to termination of breastfeeding (Chang *et al.* 2019). Break time in between working hours may be helpful to support breastfeeding mothers to find time to express their breastmilk.

Stigma of public breastfeeding

Mothers still feel uncomfortable to breastfeed in public in this 21st century. Stigma of public breastfeeding exist in this population resulting the mothers to find a closed and private place to breasted. Some chose to breastfeed in the car before heading to their destinations with their infants.

"*Of course, I feel shy to breastfeed in public. I will breastfeed my baby in the car before going out*", NBFI2 mentioned one mother.

It is clear that although breastfeeding is encouraged by family members, healthcare providers and is a normal practice of feeding a child, but stigma of breastfeeding still exist in the studied population. The inconvenience of not being able to breastfeed anytime at any location resulted in the mothers to opt for formula milk to feed their child in public.

A review on breastfeeding among overweight and obese mothers found that these group of mothers are embarrassed to breastfeed in public because of the fear of exposing their body to others (Chang *et al.* 2020). A study in Australia among breastfeeding mothers also reported that mothers often feel uncomfortable to breastfeed in public as they constantly need to avoid male gazing at them or people coming to see the baby while they are breastfeeding (Sheehan *et al.* 2019). Hence, they often cover up and this led to them feeling scared of suffocating their child or afraid that their child is not being fed properly. This discomfort remains a barrier in many countries in which social norm include them to cover themselves up properly if they breastfeed in public (Coomson & Aryeetey 2018; Sheehan *et al.* 2019). It is important to create awareness among public and breastfeeding mothers in order to provide a comfortable environment for mothers to breastfeed to support breastfeeding.

CONCLUSION

Many guidelines and policies exist on breastfeeding suggesting that people are aware of the importance of breastfeeding globally. However, facilitators and barriers to breastfeeding differ according to population. This study suggests that maternal perceived insufficient breastmilk, returning to work and being ashamed to breastfeed in public as main barriers to breastfeed in the studied population. Hence, intervention should aim on increasing knowledge of mothers on coping with issues related to breastfeeding difficulties such as how to increase or sustain milk production and increasing awareness of employers and public on supporting breastfeeding mothers.

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DECLARATION OF INTERESTS

The authors declare that they have no conflicts of interest.

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Correlation Between Nutritional Status, Cognitive Function and Daytime Sleepiness of Schoolchildren in Terengganu, Malaysia

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ABSTRACT

This cross-sectional study aimed to determine the correlation between nutritional status, cognitive function and daytime sleepiness of schoolchildren in Kuala Nerus, Terengganu. It involved 85 schoolchildren aged 9 to 12 years. Nutritional status was determined through anthropometric measurements, biochemical analyses for hemoglobin, urinalysis and urinary iodine concentration (UIC). While cognitive performance was tested using Raven's Colored Progressive Matrices (R-CPM). The daytime sleepiness of the respondents was assessed using the Pediatric Daytime Sleepiness Scale (PDSS). It was found that 16.5% of the respondents were stunted while 4.7%, 12.9% and 17.7% were obese, overweight and wasted, respectively. In addition, 37.7% of the respondents were anemic and the median UIC was 177.0 µg/l, which was categorized as normal for children. The majority (37.6%) of the respondents had an average cognitive level. Furthermore, the mean for daytime sleepiness based on PDSS score for overall respondents was categorized under an average level that was 14.1 score. In the present research, results found that all of the factors examined in this study were not correlated with respondents' cognitive function. As the majority of the children had average to low levels of cognitive functions, it is recommended to investigate further factors associated with cognitive functions and, subsequently, to design and deliver appropriate intervention.

Keywords: cognitive function, daytime sleepiness, nutritional status, schoolchildren

INTRODUCTION

Malaysian schoolchildren face malnutrition problems, including both underweight and overweight (Zainuddin *et al.* 2013). Based on the latest National Health and Morbidity Survey (NHMS-2019), it was reported that 29.8% of children 5−17 years of age are overweight (15.0%) and obese (14.8%). A decade ago, the prevalence of overweight among children 0 to 13 years old was 5.3% (95% CI: 4.5−6.2), and of obesity 8.0% (95% CI: 7.5−8.6). Because of the greater age range, the NHMS-2019 reported a higher proportion. The prevalence of underweight, stunting and thinness among schoolchildren in rural areas was higher than among schoolchildren in urban areas (Chung 2015; Kuay *et al.* 2013).

Cognitive function among primary schoolchildren might be influenced by several factors, such as gender, birth weight, children's nutritional status, parental education, household income, hemoglobin and iron status (Hamid *et al.* 2011). The differences in nutritional status in children may influence their food intake. For example, it is common that children that are overweight and obese are triggered to increase their daily food intake. Mental functions have biological roots, and the impairment of some neurochemical processes owing to a lack of trace elements may have mental effects. Thyroid hormones are essential for brain development and proper brain function throughout life, and iodine is required for their production (Abel *et al.* 2017), and its deficiency can be associated with mental retardation (Janka 2019). Furthermore, it has been reported that iodine exerts neuroprotective effects and improves intellectual performance (Martínez García *et al.* 2018), which includes cognitive function. The ability to cognitively control impulses may play a role in determining nutritional status, as negative moods have resulted in an increase of food intake that may

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lead to overweight and obesity among children (Tice *et al.* 2001). A study by Haile *et al.* (2016) found a significant correlation between height-for-age with cognitive function among children. Height-for-age represents the accumulation of dietary deprivation over time, which may have an impact on children's academic achievement (Shariff *et al.* 2000).

Previous studies reported that children with low hemoglobin (less than 12 mg/dl) had a substantially lower cognitive function score than those with normal hemoglobin (more than 12 mg/dl) status as measured by Raven's test (Hamid *et al.* 2011). In terms of iodine intake, a previous study found that one third of the world's school-age population has urinary iodine below 100 µg/l, which indicates insufficient iodine intake, while 39.9% of school-age children in South East Asia also had insufficient iodine intake (Kuay *et al.* 2013). Furthermore, Kuay *et al.* (2013) also reported that the median Urinary Iodine Concentration (UIC) of children in urban and rural areas in Terengganu was 87.7 µg/l and 72.4 µg/l, respectively, which falls into the Iodine Deficiency (ID) range. Al-Mekhlafi *et al.* (2011) reported that children who suffered from iodine deficiency were more prone to having lower cognitive function and educational achievement than children with sufficient iodine intake.

In addition to nutritional status, previous research also suggests that daytime sleepiness can have a negative impact on students' academic performance and motivation (Abdulghani *et al.* 2012). Poor sleep was significantly correlated with lower performance of cognitive functions among schoolchildren (Lam *et al.* 2011). Sleepiness defined as individual self-reported total sleep time and daytime sleepiness are associated with poor academic achievement and other negative outcomes such as unstable mood (Drake *et al.* 2003). Further studies on daytime sleepiness are suggested as it may influence cognitive performance and, in particular, memory and cognitive function (Ahmad & Bashir 2017).

Therefore, based on the knowledge acquired from previous studies regarding the malnutrition problem among children in rural Malaysia and the present of associations between nutritional status, anemia and iodine deficiencies, daytime sleepiness with cognitive function this study aimed to assess the correlations of these factors among school children in Kuala Nerus. Hence, it can provide the latest data on factors that associated with cognitive function among schoolchildren aged 9 to 12 years old as well as baseline information regarding daytime sleepiness and cognitive function among this demography which is not widely reported in previous studies especially in Malaysia. In addition, the data gained from this study can also be used as a baseline for schoolchildren aged 9 to 12 years old in other states other than Terengganu in similar situations.

METHODS

Design, location, and time

The present study was a cross-sectional study carried out in two primary schools located in Kuala Nerus, Terengganu. Two primary schools were chosen, namely, SRI Al-Amin and SK Tanjung Gelam. The respondents were sampled in both schools within a two-month period. Ethical approval for this study was obtained from the Research, Management and Innovation Centre, Universiti Malaysia Terengganu with reference number: UMT/RMIC/2-2/62 (81).

Sampling

In the present study, convenience sampling was used. There were five primary schools that had been selected in Kuala Nerus. However, through convenience sampling, only two primary schools were chosen to be involved in this study. These schools were chosen because of easy accessibility as their headmasters were willing to cooperate by giving their permission. Besides, the respondents were available during the data collection.

The criterion for inclusion of the respondents in both schools were schoolchildren aged 9 to 12 years who were approved by their parents or guardians. This age group was chosen based on a discussion with the school administration that allowed researchers to be involved with at least 9 years of children. Information consent sheets that thoroughly explain the objectives and methodology of this study were also given to the respondents and parents along with the consent sheet. The Yamane (1967) formula was applied in order to determine the sample size of this study with a 95% confidence interval. The common margin of error used in other studies ranges from 5% to 10%. Therefore, the margin of error used

in this study was in between the range, which was 7%. The size of the population of this study was 145 respondents, and on the basis of the formula, the calculated sample size was 85 respondents.

Data collection

The tools used for sampling in the present study were a questionnaire, anthropometry measurements and biochemical analyses. The questionnaire was divided into three sections: Section A contained socio-demographic information, while Section B included eight items assessing respondents' daytime sleepiness following Drake *et al.* (2003), whereby a higher score on the Pediatric Daytime Sleepiness Scale (PDSS) indicated greater levels of sleepiness. Lastly, Section C was the cognitive function test, with 36 matrices of diagrammatic puzzles of Raven's Colored Progressive Matrices (R-CPM) (Raven 2008).

Anthropometry data used for this study included body height, body weight and BMI. In this study, weight was measured using a Tanita digital body fat monitor/scale UM-026 (Tanita, UK). Body height measurements were also taken without shoes and socks (SECA, Germany). The weight and the height were measured to the nearest 0.1 kg and 0.1 cm, respectively (Hamid *et al.* 2011). Height-for-age was then categorized as tall, normal, stunted, and severe stunted while BMI-for-age was classified as obesity, overweight, normal, thin, and severe thin (WHO 2007). The WHO AnthroPlus version 1.0.3 was used to determine Z-scores for height-for-age and BMI-for-age among children.

In terms of biochemical analysis, the instrument used for testing hemoglobin concentration was a photometer called HemoCue®. The urinalysis was performed by using the Urine Analyzer PocketChem™ UA PU-4010 in order to determine the presence of bilirubin, blood, glucose, ketone, leukocyte, nitrite, pH, protein, specific gravity and urobilinogen. Only a small amount of the mid-stream of urine was needed for the test. The laboratory procedure widely used to determine the concentration of urinary iodine is based on the *Sandell-Kolthoff* reaction (Jooste & Strydom 2010).

Data analysis

In the present study, the Statistical Package for Social Sciences (SPSS version 20.0) was used to analyze the data. Spearman's Rank Order Correlation was used for non-parametric test to measure the relationship between the parameters of the test. In all analyses, $p<0.05$ was used to indicate significant outcomes.

RESULTS AND DISCUSSION

Table 1 shows the socio-demographic characteristics of children. The majority (45.9%) of respondents were aged 11 years old, all from Malay families. More than half of the children live in households that consist of four to six people. In terms of parents' employment status, most of their fathers (92.9%) were working and more than half of mothers (67.1%) were also working.

Nutritional status based on anthropometric measurement

Table 2 presents the mean height, body weight, BMI-for-age and height-for-age of the children. In terms of BMI-for-age, the majority of the respondents (56.5%) were in normal condition, while 4.7%, 12.9%, 10.6%, and 7.1% were obese, overweight, wasted and severely wasted, respectively. On the other hands, majority (83.5%) of the respondents had normal height-for-age while 9.4% and 7.1% were stunted and severely stunted, respectively.

As stunting reflects past nutrition, during their early childhood, some of these children might have experienced poor food intake and had some sort of infection (Zainuddin *et al.* 2013). In addition, they probably were consistently living in an unchanged situation due to underprivileged living conditions (Shariff *et al.* 2000). Picauly & Toy (2013) reported that the malnutrition conditions, such as stunting, experienced by school-age children will affect their ability to learn in school and, subsequently, may affect their learning achievement. Besides, it has been reported that children's sex and age affect both wasted and stunted. With increasing age, they are more prone to stunting but without wasting (Shariff *et al.* 2000).

Nutritional status based on biochemical analysis

In the present study, the majority (62.35%) of the respondents had a normal reading of hemoglobin concentration, while more than one third of the respondents were anemic. Ngui *et al.*

Table 1. Socio-demographic characteristics of respondents

Characteristics	n=85	%
Gender		
Boy	46	54.1
Girl	39	45.9
Age of child (years old)		
9	15	17.6
10	12	14.1
11	39	45.9
12	19	22.4
Median (IQR): 11 (1)		
Race		
Malay	85	100
Number of family members (including parents)		
1−3	3	3.5
4−6	45	52.9
7−9	36	42.4
≥10	1	1.2
Range: 1−10		
Median (IQR): 2 (1)		
Employment status of father		
Working	79	92.9
Not working	6	7.1
Employment status of mother		
Working	57	67.1
Not working	28	32.9

(2012) reported that anemia among adolescent is caused by physiological iron losses during menstrual bleeding in female and increased iron needs during the growth spur period.

Based on Table 3, the overall median UIC of respondents was 177.0 µg/l, which was categorized as a normal UIC for children. In general, this reflects that the children had sufficient iodine intake in their daily food intake. According to the study by Selamat *et al.* (2010) in different locations, the median UIC of children aged eight to ten years was in the normal range, in terms of Malaysian data (109 µg/l), and data for Peninsular Malaysia (104 µg/l), Sabah (150 µg/l) and Sarawak (102 µg/l). A mandatory Universal Salt Iodization (USI) has been implemented in Sarawak, Malaysia since 2008. Recently, Kuay *et al.* 2021 reported the overall median Urinary Iodine Concentration (UIC) level among the school-aged children in Sarawak was 126.0 µg/l (Interquartile Range (IQR): 71.0−200.9 µg/l) and classified as adequate iodine status.

In the present study, the median UIC for children in Terengganu (177.0 µg/l) was higher compared with the other regions, as stated above. The reason for acceptable UIC among schoolchildren in Terengganu might be due to the availability of iodine rich food sources to

Table 2. Anthropometric measurements and growth status of respondents

Parameter	Overall (n=85)
Height (cm)	140.50±8.08
Weight (kg)	35.73±10.72
BMI-for-age	
Normal	48 (56.5%)
Obese	4 (4.7%)
Overweight	11 (12.9%)
Possible risk of overweight	7 (8.2%)
Wasted	9 (10.6%)
Severely wasted	6 (7.1%)
Height-for-age	
Normal	71 (83.5%)
Stunted	8 (9.4%)
Severely stunted	6 (7.1%)

*BMI: Body Mass Index

Table 3. Hemoglobin and urinary iodine concentration

Indicators	Overall (n=85)	Range
Hemoglobin concentration (g/dl)	12.00±1.50	6.00−15.80
Normal	53 (62.35%)	
Anemic	32 (37.65%)	
Urinary iodine concentration (µg/l)	32 (37.65%)	24−245

Data are presented as mean±SD or median (IQR)

residents of the community, resulting in a higher frequency of iodine food consumption and a lower risk of iodine deficiency. A study by Kuay *et al.* (2013) showed that the median UIC of children who lived in urban areas in Terengganu was 87.7 µg/l, which falls into the iodine deficiency range. However, in the present study, the median UIC of children who lived in Terengganu was categorized as a normal UIC for children. Thus, the consumption of iodine among children in this state has improved over the last five-year period.

In terms of urinalysis, all of the respondents had no glucose in their urine. The majority (98.82%) of respondents had no protein in their urine. According to Simmerville *et al.* (2005), protein is not normally found in urine, as most proteins are too big to diffuse out of the glomerular filtrate. All of the respondents had normal urine pH, which was between 4.5 to 8.0 and this indicates that the respondents did not have UTI or calculi. Furthermore, the majority of the respondents (76.47%) had normal Urinary Specific Gravity (USG) and for some respondents that showed high USG, they might be suffering from dehydration due to inadequate fluid intake in school. Some of the factors that raise specific gravity include dehydration, diarrhea, emesis, UTI and proteinuria as well as water restriction (Simmerville *et al.* 2005). However, 16.47% of the respondents showed positive blood results in their urine. The abnormal blood found in urine among male respondents might be due to infection, whereas positive blood results among female respondents were probably because they were undergoing menstruation when the test was carried out. Lastly, the majority (96.47%) of respondents had negative leukocyte esterase in their urine, while those who showed positive results were suspected of having an infection.

Raven cognitive function

As shown in Table 4, the majority (37.6%) of the respondents had an average cognitive level, followed by high average, low average and extremely low cognitive levels with 25.9%, 12.9% and 10.6%, respectively. The mean R-CPM score for overall respondents was 93.5, in the range of 9.0 to 135.0 of total scores. The R-CPM test depends on non-verbal logical reasoning.

In addition, a recent study reported that varied improvements in cognitive skills (as

Table 4. Cognitive function level of respondents

Parameter	Overall (n=85)	Mean score	Range score
Cognitive function level		93.5±20.2	9−135
Extremely low	9 (10.6%)		
Borderline	8 (9.4%)		
Low average	11 (12.9%)		
Average	32 (37.6%)		
High average	22 (25.9%)		
Superior	2 (2.4%)		
Very superior	1 (1.2%)		

measured using R-CPM) were observed for different variables in both the intervention and control groups of preschool children with no robust evidence for physical activity-intervention-related improvements (Jaksic *et al.* 2020).

Daytime sleepiness

Daytime sleepiness, including sleep continuity, how rested an individual feels upon waking up, and how alert one is during the day, has an important effect on educational outcomes in children (Blunden *et al.* 2018). In addition, sleep can also affect young adolescents' mental, social, physical and psychological components (Blunden *et al.* 2018). However, lack of sleep time and daytime sleepiness among school-aged children were linked with low scores in neuropsychological attention studies (Zainuddin *et al.* 2013). A meta-analysis investigating the effects of school start times on sleep and daytime sleepiness has discovered that later school start times result in longer sleep durations and reduced daytime sleepiness in adolescents (Bowers & Moyer 2017). In a systematic review, Meyer *et al.* (2017) found a moderate use of the Paediatric Daytime Sleepiness Scale (PDSS) to assess daytime sleepiness. This tool provides for the tracking of factors that influence children's and adolescents' excessive daytime drowsiness. The mean score for daytime sleepiness based on the (PDSS) for overall respondents was 14.1 with a range of 5 to 25 scores. It was categorized under average level scores. On the other hand, higher PDSS scores indicate a higher level of sleepiness associated with the individual's total sleep time (Drake *et al.* 2003). Based on a study by Drake *et al.* (2003), the mean score PDSS for overall respondents was 15.3, almost similar to the mean score of the present study, which was at 14.1. Another study by Perez-Chada *et al.* (2007) reported that the PDSS (mean score: 15.7) was robustly and significantly associated with academic failure.

Correlation between nutritional status and daytime sleepiness with cognitive function among respondents

The present study found no significant correlation between height-for-age and BMI-for-age with cognitive function. The results also demonstrated no significant correlations between hemoglobin and urinary iodine concentration with cognitive function. Lastly, there was also no significant correlation between daytime sleepiness and cognitive function, as shown in Table 5.

These findings are in agreement with a study by Hamid *et al.* (2011) that reported height-for-age z-scores are not apparently correlated with cognitive tests. In contrast, Haile *et al.* (2016) found a significant correlation ($r=0.38$; $p=0.001$) between children's height-for-age and cognitive function. Another study also reported that brain functions including cognition, memory and locomotors skills are influenced by under-nutrition (Ranade *et al.* 2008). A study by Haile

Table 5. Correlation between nutritional status and daytime sleepiness with cognitive function

Parameter	Cognitive function	
	r	p
BMI-for-age	-0.116	0.292
Height-for-age	-0.021	0.850
Hemoglobin concentration	-0.170	0.120
Urinary iodine concentration	0.076	0.489
Daytime sleepiness	-0.017	0.876

*BMI: Body Mass Index

et al. (2016) reported no significant correlation between BMI-for-age with cognition, similar to the present findings.

In contrast, a study by Hamid *et al.* (2011) reported that hemoglobin concentration was significantly correlated with cognitive function (r=0.19; p=0.006). In their findings, it was reported that children with iron deficiency had significantly lower cognitive function test scores than their normal hemoglobin counterparts (Hamid *et al.* 2011). Another study by Al-Mekhlafi *et al.* (2011) among children in rural areas revealed that those suffering from iron deficiency anemia were more prone to having lower cognitive function and academic performance than healthy children. Across past studies, it has been conclusively reported that iron deficiency anemia delays the psychomotor development and impairs the cognitive scores of pre-school and school-age children in Thailand, Indonesia and Zaire (Al-Mekhlafi *et al.* 2011). Several studies have linked iron deficiency anemia to cognitive impairment, psychomotor disorders, and behavioral problems in children of all ages. Disorders of attention, emotion, intelligence, and sensory perception are the most common cognitive functions impaired by iron deficiency. These long-term impacts can have a negative impact on learning ability and professional ability acquisition (Radlowski & Johnson 2013; Jáuregui-Lobera 2014).

Despite the well-documented importance of adequate iodine intake for early-life neurodevelopment, Kippler *et al.* (2016) found no association between the mothers' urinary iodine concentration (median 172 µg/l) and children's general cognitive score, even when comparing children born to mothers with an inadequate iodine status (urinary concentrations less than 150 µg/l) (36% of the women) to those born to mothers with an adequate iodine status (urinary iodine concentration of more than 150 µg/l). This might be due to the iodine deficit that was discovered was not be frequent or severe enough to impact neurodevelopment.

The present study also shows no correlation between daytime sleepiness and cognitive function. This finding also was not in agreement with Lam *et al.* (2011), who reported that daytime sleepiness was significantly correlated with cognitive function among schoolchildren. Another study by Moreau *et al.* (2013) also reported that lack of sleeping time among school-aged children is associated with lower performance on neuropsychological tests of attention. Drake *et al.* (2003) stated that sleepiness would be related to individual total sleep time and quality that would be associated with poor academic achievement and other negative outcomes, such as poor mood.

CONCLUSION

In conclusion, the majority of the respondents had normal growth for both the indicators of height-for-age and BMI-for-age. Based on urinary iodine analysis, the results indicate that they received sufficient micronutrients such as iodine in their daily food intake. The majority of them had average cognitive function levels. In the present study, there was no significant correlation (p>0.05) between anthropometric indices (height-for-age and BMI-for-age), hemoglobin concentration, urinary iodine concentration and daytime sleepiness with children's cognitive function.

The present study provides the latest data about the correlation between nutritional status, daytime sleepiness and cognitive function among schoolchildren in Kuala Nerus. This data is useful for future reference, especially the data about daytime sleepiness, as it was not widely investigated in the previous studies especially in Malaysia. As the majority of the children had average to low levels of cognitive functions, it

is recommended to investigate further factors associated with cognitive functions and, subsequently, to design and deliver appropriate intervention.

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DECLARATION OF INTERESTS

The authors confirm that there are no known conflicts of interest associated with this publication.

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