Effects of Duration of Breastfeeding During Infancy on Vascular Dysfunction in Adolescents

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ABSTRAK

Tujuan: *mencari hubungan antara lama pemberian ASI pada masa bayi dengan karakteristik vaskular sebagai faktor risiko kardiovaskular pada anak remaja.* ***Metode:*** *Studi retrospektif kohort terhadap remaja berumur 15-18 tahun. Lama pemberian ASI diperoleh dengan kuesioner terhadap orangtua yang dikategorikan: 0-<2 , 2-<4, 4-<6, 6-,12 dan > 12 bulan. Luaran yang dinilai adalah "flow mediated dilation" (FMD), ketebalan tunika intima media (CIMT) dan ukuran antropometri. Dilakukan analisis statistik menggunakan regresi linear dan MANOVA general linier model dengan faktor risiko kardiovaskular sebagai variabel dependen dan durasi pemberian ASI sebagai variabel independen dan dengan koreksi terhadap faktor perancu (confounders).* ***Hasil:*** *terdapat 285 subjek penelitian berumur 15-18 tahun. Durasi pemberian ASI 4-<6 bulan ternyata mempunyai asosiasi dengan ketebalan tunika intima media (CIMT) dan lebih menonjol bila disesuaikan dengan jenis kelamin dan terpajan dengan rokok pasca-natal (beda rerata 14,28 mikrometer, p=0,045). Tidak ada hubungan bermakna antara durasi pemberian ASI dengan flow mediated dilation (FMD).* ***Kesimpulan:*** *Lama pemberian ASI 4-<6bulan berhubungan dengan ketebalan tunika intima media karotis (CIMT) sehingga mempunyai efek proteksi terhadap terjadinya penyakit kardiovaskular. Namun demikian hubungan durasi pemberian ASI dengan FMD pada remaja belum jelas.*

Kata kunci: *durasi pemberian ASI, fungsi dan struktiur vaskular, risiko kardiovaskular.*

ABSTRACT

Aim: *to investigate the effect of breastfeeding duration on vascular characteristics in adolescence.* ***Methods:*** *we conducted a retrospective cohort study on adolescents aged 15-18 years old. Breastfeeding duration was inquired using a questionnaire filled by parents and categorized into 0-<2, 2-<4, 4-<6, 6-<12, and >12 months. Outcomes assessed were flow mediated dilation (FMD), carotid intima media thickness (CIMT), anthropometrics. Analysis was done using linear regression and MANOVA general linear model with cardiovascular risk factors as the dependent variables and breastfeeding duration as the independent variable with further adjustment for confounders.* ***Results:*** *285 subjects aged 15-18 years were enrolled. Breastfeeding duration of 4-<6 months was associated with thinner CIMT and the effect was more prominent after adjustment for gender and postnatal tobacco exposure (mean difference=24.28 micrometer, p=0.045). No statistically significant association was found with FMD.* ***Conclusion:*** *breastfeeding duration of 4-<6 months is associated with thinner IMT and thus has a protective effect on the development of cardiovascular disease. However the association with FMD in adolescence is less clear.*

Key words: *breastfeeding duration, vascular structure and function, cardiovascular risk.*

INTRODUCTION

Cardiovascular disease (CVD) remains as the number one cause of death worldwide.[1] It is estimated that 17.3 million deaths occur each year due to CVD and 48% of deaths related to non-communicable diseases are caused by CVD.[2] The prevalence is even increasing in developing countries including in Indonesia,[3] where cerebrovascular and coronary heart diseases are the first and second most causes of death, respectively.

It is widely recognized that the development of CVD has started in early life.[4,5] Increased cardiovascular risk factor levels have also been found in children and adolescents. A study in Jakarta showed that 16.1% of young adults had hyperlipidemia, 32.6% were overweight or obese, and 48.8% had hypertension.[6] Several studies also found a high prevalence of hypercholesterolemia (25-33%) among Indonesian schoolchildren. Autopsy studies have also shown that atherosclerosis is already found in young adults. Moreover, fatty streaks, the initial lesions of atherosclerosis, have been found in children less than one year old and its occurrence increases with age, which is 20% in children aged 15-19 years and 40% in the age of 30-34 years.[7]

It has been suggested that changes in intima media thickness (IMT) and endothelial dysfunction are early signs of atherosclerotic plaque formation.[8,9] Endothelial dysfunction can be assessed by the flow mediated dilation technique. The measurement uses ultrasound to detect changes in brachial vessel diameters after a shear stress is applied. Intima media thickness reflects changes in vascular structure and has been considered as a strong predictor of CVD in adults. Other biomarkers considered to be associated with early vascular injury are high-sensitive C-reactive protein (hsCRP) and the vascular cell adhesion molecule (VCAM).[10,11]

Various factors have been implicated to be involved in atherosclerosis formation. These include nutrition, physical activity, smoking exposure, obesity, hypertension, diabetes mellitus. In particular, nutrition in infancy has been suggested to play a role in the development of CVD.[12] Previous studies have indicated a protective effect of breastfeeding on CVD, in which adults who used to be breastfed have better arterial function than those who were formula fed.[13] It is suggested that the long-chain polyunsaturated fatty acid (LC-PUFA) contained in breastmilk increased vasodilator production and release, such as nitric oxide.[14] LC-PUFA may also have anti-atherosclerotic properties through suppressing pro-inflammatory cytokines. It has also been reported that breastfeeding of at least 3 months has, at least in adults, a positive effect on cardiovascular biomarkers, such as lower total/LDL cholesterol and high sensitive C-reactive protein (hs-CRP) levels.[15]

Although several studies show that breastfeeding has a preventive effect on CVD, there are also studies revealing inconsistent results, in which longer duration of breastfeeding is associated with decreased arterial function and increased blood pressure in later life.[16] Moreover, the optimal breastfeeding duration in relation to cardiovascular risk is largely unknown. Since early detection of atherosclerosis risk factors at a young age would enable early intervention to prevent cardiovascular disease in adulthood, we aimed to evaluate the effect of breastfeeding duration on vascular characteristics and other cardiovascular risk factors in adolescents.

METHODS

We conducted a retrospective cohort study on adolescents aged 15-18 years old from October 2012 until March 2013. Subjects were recruited from a senior high school (SMAN 1 Budi Utomo) in Jakarta, Indonesia. The number of subjects were calculated using the formula of mean difference with the type I error of 5% and type II error of 10% The minimal mean diffrence of CIMT which is clinically significant was 0.45 and standard deviation of 0,40 mm. The minimal subjects of each category duration of breastfeeding was 19 subbjects. Only adolescents whose mothers were able to recall the breastfeeding duration were enrolled. We excluded subjects who had an acute fever or illness during examination. Parental informed consent was obtained for all subjects and the study has been approved by the Ethics Committee of Faculty of Medicine Universitas Indonesia.

Two weeks before enrolment, a health information session was arranged in school to explain the importance of preventing CVD at a young age and to invite students to participate in the study. Of 300 students invited, 15 children rejected, mainly because of refusal to the blood sampling procedure. After consent had been obtained, a questionnaire was applied to parents to inquire for breastfeeding duration, which was further categorised into 0-<2, 2-<4, 4-<6, 6-<12, and >12 months. Using the questionnaire, we also obtained information about age, gender, socioeconomic level, mothers' age, and smoking exposure. We performed complete physical examination including subjects' blood pressure, body weight and height, as well as waist and hip circumference. Blood sampling was taken after a 12-hour fasting to measure total/HDL/LDL cholesterol, glucose, hs-CRP, and VCAM levels.

After resting for 10 minutes, subjects underwent blood pressure, FMD, and IMT examinations in supine position. To measure FMD, we first measured the brachial artery diameter using a vascular ultrasound (General Electric Logic E) with a 7-12 MHz transducer. Afterwards, using a sphygmomanometer, we applied pressure of 200 mmHg or approximately 50 mmHg above systolic pressure to occlude the brachial artery for 1 (FMD1) and 4 minutes (FMD4). After pressure was released, diameter measurement was repeated to obtain the ratio of vascular diameter after occlusion to that of before occlusion. We also used the ultrasound device to measure CIMT. All ultrasound examinations were done by an ultrasound technician blinded to data on breastfeeding duration.

Data were firstly described in a baseline table and presented as mean, median, or proportion as appropriate. To evaluate the association between breastfeeding duration and vascular characteristics as well as other cardiovascular risk factors, we initially performed univariable analysis using one-way ANOVA with breastfeeding duration categories and potential confounders as the independent variables and CIMT, FMD, as the dependent variables. Potential confounders were taken into the multivariable model.

Multivariable analysis was done using MANOVA general linear model with breastfeeding duration dichotomised into >4-6 months and other than >4-6 months as independent variable, while, CIMT, FMD1, hs-CRP taken simultaneously as the dependent variables due to possible interactions between these outcomes. The analysis was adjusted to gender and postnatal smoking exposure as both variables are likely to be associated with breastfeeding duration and cardiovascular risk factors. We particularly focus on breastfeeding duration of >4-6 months since it is the category that showed an association with vascular characteristics on univariable analysis and was in line with global recommendation of breastfeeding duration and also in the multivariate analysis the group of 4-6 months duration of breastfeeding tends to have a good results.

RESULTS

Two hundred and eighty five senior high school students were enrolled. Subjects' characteristics based on breastfeeding duration categories are shown in **Table 1**. There was a statistically significant difference in body weight across breastfeeding categories, in which prolonged breastfeeding was associated with smaller body weight (p=0.02). There was also a significant difference in mother educational level across BF duration categories. More mothers with low-to-middle education gave prolonged breastfeeding (more than 12 months) as compared to other categories (p=0.006). We found no significant differences in gender, birth weight, height, social economy status, and subjects' exposure to parental smoking (before or after birth) based on breastfeeding categories.

Results of univariable analysis of the association between breastfeeding categories and vascular characteristics as well as other cardiovascular risk factors were shown in **Table 2**. With increasing breastfeeding duration, blood pressure and CIMT decreased although not statistically significant. Children with breastfeeding duration of 4-<6 months had the thinnest IMT compared to others. In

Table 1. Baseline characteristics by breastfeeding duration

Variables	Breastfeeding duration (months)						p value
	0 - <2 (n=27)	2 - <4 (n=42)	4 - <6 (n=25)	6 - <12 (n=54)	>12 (n=137)	Total	
Male sex, n (%)	8 (30)	18 (43)	11 (44)	22 (41)	39 (28)	98 (34)	0.22*
Birth weight,grams, n (%)							
- < 2500	3 (11)	3 (7)	2 (8)	2 (4)	9 (7)	19 (7)	0.74#
- > 2500	24 (89)	39 (93)	23 (92)	52 (96)	128 (93)	266 (93)	
Age, year, mean (SD)	15.5 (0.9)	15.6 (1.0)	15.6 (1.0)	15.4 (0.8)	15.7 (1.0)	15.6 (1.0)	0.61**
Body weight, kg, mean (SD)	59.9 (11.4)	60.9 (16.1)	58.3 (14.4)	57.5 (13.6)	54.2 (11.3)	56.7 (13.0)	0.02**
Height, cm, mean (SD)	159.3 (6.5)	162.2 (8.1)	161.3 (8.5)	161.1 (8.2)	159.2 (7.8)	160.2 (7.9)	0.17**
Current age mother (years)							
- 30-40	8 (30)	8 (19)	4 (16)	9 (17)	35 (25)	64 (23)	0.77*
- > 40	19 (70)	34 (81)	21 (84)	45 (83)	102 (75)	221 (77)	
Socioeconomic status, n (%)							0.20*
- Low	8 (30)	10 (24)	7 (28)	13 (24)	49 (36)	87 (30)	
- Middle	14 (52)	28 (67)	16 (64)	35 (65)	80 (58)	173 (61)	
- High	5 (18)	4 (9)	2 (8)	6 (11,1)	8 (6)	25 (9)	
Mother education, n(%)							0.006*
- Low	1 (4)	2 (5)	2 (8)	5 (9)	16 (12)	26(9)	
- Middle	10 (37)	17 (40)	9 (36)	21(39)	68 (50)	125 (44)	
- High	16 (59)	23 (55)	14 (56)	28 (52)	53 (39)	134 (47)	
Prenatal parental tobacco exposure, n (%)	12 (44)	14 (33)	9 (36)	22 (41)	64 (47)	164 (58)	0.25*
Postnatal parental tobacco exposure, n (%)	13 (48)	14 (33)	9 (36)	22 (41)	65 (47)	123 (43)	0.31*

Note: *Chi-square test, **ANOVA test, #Fischer Exact test

further multivariable analysis we dichotomised breastfeeding duration to obtain more number of subjects within each group. **Table 3** shows the general linear model analysis with breastfeeding duration dichotomised as 4-<6 months and other than 4-<6 months as independent variable. There were statistically significant associations between breastfeeding duration of 4-<6 months and thinner CIMT both in crude and adjusted analysis to gender and postnatal parental tobacco exposure (p=0.045).

DISCUSSION

This study provides further insight about factors associated with the development of atherosclerosis that starts early in life. We found that adolescents who had been breastfed for 4-<6 months have thinner CIMT compared to others. They also tended to have larger FMD but the association was not statistically significant.

Flow mediated dilation (FMD) is one of the sensitive tools for detection of early dysfunction of the vascular endothelium. This study showed that there was no significant difference in 1 minute and 4 minute FMDs across BF duration categories although FMD1 and FMD4 tended to be higher in the adolescents who had been breastfed for 4-<6 months compared to other categories. In terms of CIMT, we found that children who used to receive breastfeeding for 4-<6 months had thinner CIMT compared to other groups of BF duration. Our findings were slightly different from a previous study in Finland, which showed that FMD was highest

Table 2. Cardiovascular risk factors based on breastfeeding duration categories (univariable analysis)

Variables	Breastfeeding duration (months)					p value
	0 - <2 (n=27)	2 - <4 (n=42)	4 - <6 (n=25)	6 - <12 (n=54)	>12 (n=137)	
Vascular characteristics and biomarkers, mean (SD)						
FMD 1 minute (%)	9.2 (7.1)	9.2 (6.1)	10.9 (6.7)	9.2 (5.9)	9.5 (5.9)	0.80
FMD 4 minutes (%)	15.9 (7.1)	15.3 (8.4)	15.6 (8.1)	14.6 (7.5)	15.0 (6.9)	0.95
CIMT (µm)	442.2 (67.8)	445.7 (63.0)	412.8 (56.1)	447.6 (77.6)	429.3 (67.6)	0.15
Systolic BP (mmHg)	119.8 (15.5)	122.5 (16.2)	114.1 (15.8)	118.4 (18.5)	115.2 (13.8)	0.06
Diastolic BP (mmHg)	70.9 (7.7)	72.7 (10.9)	68.5 (15.6)	70.8 (11.7)	68.9 (11.7)	0.37

Table 3. Association between breastfeeding duration of 4-<6 months and vascular characteristics/cardiovascular risk factors

Variables	BF duration (months)		p value	
	4 - <6 (n = 25)	Other duration (n = 260)	Unadjusted	Adjusted*
Vascular characteristics and biomarker, mean (SD)				
FMD 1 min (%)	10.9 (6.7)	9.4 (6.0)	0.22	0.230
FMD 4 min (%)	15.6 (8.1)	15.1 (7.2)	0.72	0.700
CIMT (µm)	412.8 (56.1)	437.1 (69.2)	0.09	0.045
Systolic BP (mmHg)	114.1 (15.8)	117.5 (15.6)	0.30	0.130
Diastolic BP (mmHg)	68.5 (15.6)	70.1 (11.2)	0.52	**

**: Exclude from multivariate analysis
Multaivariat analysis Adjusted to gender and postnatal parental tobacco exposure

in children who had exclusive breastfeeding compared to those formula-fed, which was 7.2 (SD 4.0) % versus 5.9 (3.4) %, but it showed no difference in CIMT between the groups.[17,18] However that study did not evaluate the effect of BF duration as our study did. An other study in England, also found that CIMT of adults aged 32-88 years old who used to be breastfed was thinner than those who were formula-fed. However, that study did not provide evidence regarding the dose-response effect of breast milk on vascular structure and function.[13]

Evelein et al. reported the effect of breastfeeding on CMIT of 5 year old children. In this prospective cohort study, subjects who received breastfeeding for 3-6 months had CIMT 21.1 micrometer (95% CI 5.0; 37,2; p=0.01) thicker compared to those who were formula-fed, which may be due to higher cholesterol content of breastmilk than formula milk.[19] However, it was suggested that this was only a temporary condition, in which CIMT of breastfed children will later decrease in the adolescence and adulthood. This phenomenon is called "nutritional programming effect" where the cholesterol metabolism changes over time due to high cholesterol exposure in breastmilk.

In our study, blood pressure tended to be lower in adolescents with BF duration of 4-<6 months compared to other categories, but the difference was not statistically significant. There are some mechanisms by which breastmilk influences blood pressure, which are lower sodium intake and increased LC PUFA intake. LC-PUFA is an important component of vascular endothelial membrane, which protects against hyperinsulinism and insulin resistance. The association between breastfeeding and blood pressure in elderly were reported in meta-analysis by Owen et al.[20] and Martin et al.[21] that subjects who had exclusive breastfeeding will have a 1.1 (95%CI -1.79; -0.42) mmHg decrease in blood pressure compared to those who used to be formula-fed.

There were some limitations of this study. This was a retrospective cohort study involving recall of breastfeeding duration, which may cause misclassification in breastfeeding duration. Breastfeeding duration recall has been reported to be accurate until 17-20 years.[22] The other limitation of this study was that we did not perform diet analysis as diet may have influenced fat and sugar intake and thus cardiovascular homeostasis. However, blood sampling was done after subjects fasted for 12 hours. There are some confounding factors that may alter cardiovascular risk factor level such as gender, tobacco exposure, all of which has been adjusted in statistical analysis.

To our knowledge, this was the first published study investigating the effects of breastfeeding duration on cardiovascular risk factors in adolescents. The results of this study provide additional knowledge on the benefits of breastfeeding, thus it may promote breastfeeding practice, which also has been recommended globally. Since the subjects of this study have similar characteristics with most of the community in terms of socio-economic status, level of parents education, the results of this study could be applied to most adolescents in the country.

CONCLUSION

Adolescents who used to receive breast milk for 4-<6 months have thinner CIMT compared to those with breastfeeding duration of less than 4 months or over 6 months. However the association with flow-mediated dilation (FMD) and other cardiovascular risk factors seems to be more subtle.

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REFERENCES

1. World Health Organization (WHO). Global atlas on cardiovascular disease prevention and control. Geneva: WHO;2011.
2. Poulter N. Global risk of cardiovascular disease. Heart. 2003;89 (suppl 2):2-5.
3. Basic Health Research 2010, Ministry of Health, Republic of Indonesia.
4. Virmani R, Robinowitz M, Geer JC, Breslin PP, Beyer JC, McAllister HA. Coronary artery atherosclerosis revisited in Korean war combat casualties. Arch Pathol Lab Med. 1987;111:972-6.
5. Berenson GS, Wattigney WA, Tracy RE. Atherosclerosis in the aorta and coronary arteries and cardiovascular disease risk factors in person aged 6-30 years and studied at necropsy (The Bogalusa Heart Study). Am J Cardiol. 1992;70:851-8.
6. Djer MM. Prevalence of obesity in school-age children and associated factors [thesis]. Jakarta: Department of Child Health, Faculty of Medicine, University of Indonesia; 1998. Article in Indonesian.
7. Tanaka K, Masuda J, Umamura T. A nation-wide study of atherosclerosis in infants, children, and young adults in Japan. Atherosclerosis. 1988;72:143-56.
8. Bonetti PO, Lerman LO, Lerman A. Endotelial dysfunction: a marker of atherosclerotic risk. Arterioscler Thromb Vasc Biol. 2003;23:168-75.
9. Oren A, Vos LE, Uiterwaal CSPM, Grobbee DE, Bots ML. Cardiovascular risk factors and increased carotid intima media thickness in healthy young adults. Arch Intern Med. 2003;163:1787-92.
10. Zemecke A, Weber C. Inflammatory mediators in atherosclerotic vascular disease. Basic Res Cardiol. 2005;100:93-101.
11. Fragakis N, Ioannidou E, Bounda A, Theodoridou S, Klonizakis P, Garipidou V. Increased level of proinflammatory cytokines in children with family history of coronary heart disease. Clin Cardiol. 2010;8:501-5.
12. Singhal A. Early nutrition and long-term cardiovascular health.Nutr Rev. 2006;64:S44-8.
13. Martin RM, Ben-Shlomo Y, Gunnel D, Elwood P, Yarnell JWG, Smith GD. Breastfeeding and cardiovascular disease risk factors, incidence and mortality: the Caerphilly study. J Epidemiol Community Health. 2005;59:121-9.
14. Engler MM, Engler MB, Kroetz DL, Boswell KD, Neeley N, Krassner SM. The effects of diet rich in docosahexainoic acid on organ and vascular fatty acid composition in spontaneously hypertensive rats. Prostagland Leukot Essent Fatty Acids. 1999;61:289-95.
15. Ravelli AC, van der Meulen JH, Osmond C, Barker DJ, Bleker OP. Infant feeding and adult glucose tolerance, lipid profile, blood pressure, and obesity. Arch Dis Child. 2000;82:248-52.

16. Leeson CPM, Kattenhorn M, Deanfield JE, Lucas A. Duration of breastfeeding and arterial distensibility in early adult life: population based study. BMJ. 2001;322:643-4.
17. O'Tierney, Barker DJP,Osmond C, Kajantie E, Erickson JG. Duration of breastfeeding and adiposity in adult life. J Nutr. 2009;139:422S-5S.
18. Jarvisalo MJ, Hutri-Kahonen N, Juonala M, et al. Breastfeeding in infancy and arterial endothelial function later in life: the cardiovascular risk in young Finns study. Eur J Clin Nutr. 2009;63:640-5.
19. Evelein AVM, Geerts CC, Visseren FLJ, et al. The association between breastfeeding and the cardiovascular system in early childhood. Am J ClinNutr. 2011;110:1-7.
20. Owen CG, Whincup PH, Odoki K, Gilg JA, Cook DG. Infant feeding and blood cholesterol; a study in adolesecents and a systematic review. Pediatrics. 2002;110:597-608.
21. Martin RM, Ben-Shlomo Y, Gunnell D. Breastfeeding and cardiovascular disease: risk factors, incidence and mortality: the Caerphilly study. J Epidemiol Comm Health. 2005;59:121-9.
22. Natland ST, Anderson LF, Nilsen TIL, Torsmo S, Jacobsen GW. Maternal recall of breastfeeding twenty years after delivery. BMC Med Res Method. 2012;12:179.