The Effect Of Extract Moringa Oleifera Leaves Plus Royal Jelly On Hematocrit Level Of Anaemic Pregnant Women In Takalar District

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Abstract: Goal: To know effect of Moringa oleifera leaves plus royal jelly capsule supplementation on haematocrit level of anaemic pregnant woman in Takalar.

Method of study used was Experimental with randomized double blind controlle desian. Subject of the study was anaemic pregnant women in 2^{nd} and 3^{rd} trimester with gestational age of 20-32 weeks pregnancy as much as 63 samples which were divided into 3 groups namely 21 samples of Moringa leaves extract plus royal jelly (MLER) group, 21 samples of Moringa leaves extract (MLE) group, and 21 samples of placebo. Supplementation was given once a day for 60 days. Data was analysed using paired sample T-test and one way anova test.

Result: Based on the characteristics of the subjects, the age of the respondents, education, occupation, income and parity were evenly distributed or not was differrent in the three intervention groups. The highest increase of Haematocrit level after intervention was happened in group who consumed Moringa leaves extract plus royal jelly rather than Moringa leaves extract group and placebo group.

Conclusion: Moringa leaves extract plus royal jelly capsule, Moringa leaves extract capsule, and pacebo can increase haematocrit level, so this supplementation can be given to anaemic pregnant woman.

Keyword: Moringa leaves extract, Royal Jelly, Haematocrit level, Anaemic pregnant woman.

1. INTRODUCTION

Anaemia is a condition which haemoglobin level below 11 gr%. Anaemia is often occurred in pregnancy because of iron deficiency. Iron deficiency anaemia is a condition where iron is deficit which causes erythrocyte reduction. Iron deficiency anaemia in pregnancy can cause big impact such as haemorrhagic, prematurity, mother and infant mortality, and low birth weight¹. Anaemia in pregnancy is national problem because it shows social economic prosperity value of society, and its effect on human resource quality. Anaemia in pregnancy is called "potential danger to mother and child" so, anaemic pregnant woman needs special and serious attention from all connected parties in primary healthcare².WHO stated that there were 409-595 million 15-49 year old anaemic women in six countries, namely Africa America, Asia, Europe, East Mediteran, and West Pacific. Prevalence of anaemia in 15-45

year old women in Asia is 191 million women and Indonesia is in the 8th from 11th in Asia after Sri Lanka with prevalence of anaemia as many as 7,5 million in 10-19 year old women³. Based on Riskesdas 2018 there was increase of anaemic pregnant proportion in 2013 as much as 37,1% became 48,9% in 2018, while pregnant women who consumed Iron supplementation <90 tablets were 61,9%⁴. Prevalence of anaemia in pregnant woman in North Sulawesi in 2015 were 23.478 women (98,49%), while pregnant women with anaemia in Takalar in 2017 were 142 women and increased into 542 women in 2018. Prevalence of anaemic pregnant woman in South Sulawesi exceeds national rate and classified as serious problem⁵.

Anaemia can be overcome if haemoglobin (Hb) level is corrected. Iron supplementation is not definitely beneficial in increasing Hb level in body even less if it is not consumed with additional intake. Iron will form Hb if pregnant woman consumes enough protein. So, to overcome anaemia, pregnant woman needs iron supplementation and enough protein. Besides, pregnant woman also needs enough vitamin C intakes to prevent anaemia. Vitamin C helps absorption of iron in body. Iron is needed during pregnancy for foetus, placenta, and raising of erythrocyte to cover iron necessity, expansion of erythrocyte depends on iron flow from spare, diet and iron supplementation, total necessity of iron during pregnancy is about 1000 mgs.

Good intake during pregnancy is important because by consuming many macronutrient and micronutrient that is beneficial to fulfil additional nutrient needs during pregnancy. Nutrient status of pregnant woman is influenced by intake quantity of energy or calorie, protein, iron, carbohydrate, vitamin A, folic acid, calcium, iodine and other nutrients. In all of country, pregnant woman and children under 5 years old is on high risk of micronutrient deficiency. Deficiency of iron, iodine, folic acid, vitamin A, and zinc is the widest micronutrient deficiency and common contributor of bad growth, intellectual interruption, perinatal complication, and increase of morbidity and mortality⁶. Moringa leaves has many nutrients such as vitamin C which is equal to vitamin C in 7 oranges, vitamin A which is equal to vitamin A in 4 carrots, calcium which is equal to calcium in 4 glasses of milk, potassium which is equal to potassium in 3 bananas, and protein which is equal to calcium in 2 yogurts, vitamin D, vitamin E, vitamin K, biotin and folic acid. Phenol contained in Moringa leaves is 1,6% while the leaves itself is 3,4%. Besides, it also contains of ascorbic acid, flavonoid, phenoid and caratenoid of Moringa as antimicrobial and high antioxidant. The substances were needed because pregnant woman with iron deficiency anaemia can cause growth restriction, whether body cells or brain cells and decrease immunity and cause infection. Besides, indicator of iron deficiency anaemia is level of haemoglobin, ferritin, erythrocyte, and haematocrit.

Haematocrit is percentage of blood volume which consists of erythrocyte. Anaemic pregnant woman is marked by decrease of haematocrit level because of the increase of blood volume and unbalanced increase of erythrocyte production in pregnancy, so blood volumes increase for about 42% while increase of erythrocyte is about one third of it so it causes physiology haemodilution which causes decrease of haemoglobin and haematocrit level. Haematocrit and haemoglobin level of pregnant woman will decrease until its peak in late 2nd trimester of pregnancy. Study by Mastiaji Bekti 2001 which was conducted to 2nd and 3rd trimester. With enough diet, haemoglobin level will approximate initial level in early pregnancy. But if iron and supplementation back up is not enough, it can decrease haemoglobin and haematocrit level. So, it needs multidimensional, coordinated, and continuous strategy to overcome micronutrient deficiency. It needs short term and long term solution⁶. Moringa oliefera leaves extract plus royal jelly is food material which is common in area which has big potential to become herbal supplement material. It needs Moringa leaves extract plus

royal jelly as pregnant woman supplementation to increase haemoglobin, erythrocyte, and haematocrit level. Royal jelly is a product of honey bee cultivation which is related to bee's food that is pollen and nectar as main material of royal jelly production⁸. Royal jelly contains water (50%-60%), protein (18%), carbohydrate (15%), lipid (3%-6%). Besides, royal jelly contains bioactive compound such as 10-hidroksi-2 decenoic (HAD), which has benefit as immunomodulator, protein, adenosine monophosphate (AMP), adenosine, acetylcholine, polyphenol and hormone such as testosterone, progesterone, prolactin, and estradiol. Royal jelly also contains essential amino acid which is needed as many as 29 amino acid and its derivate, aspartate acid, and glutamate acid ⁹.

Studies about effect of Moringa oliefera on haematocrit which was done by Rahma Maharsi et.al 2018 showed that combination of Sauropus androgynous leaves and Moringa leaves increased haematocrit level of female wistar grove mouse¹⁰. The study divided 54 male mice into 4 groups that were distillated water control group, and 3 treatment groups which were given aqueous Moringa leaves extract oral for 21 days. There was significant increase of haematocrit level in 400mg/kg group $(45,3\pm1,6\%)^{11}$. Parallel of study by Lutfiyah Fifi 2011 that showed that Moringa leaves powder in 1,2, and 3 dose influenced representation of samples' haematology. Nutrient composition of NTB Moringa leaves powder in Africa countries. In addition to protein, components which have role in increasing Haemoglobin, erythrocyte, albumin, Iron and haematocrit¹².

Many of the studies showed that Moringa leaves extract plus royal jelly supplementation had never been done before, so this study aimed to know effect of Moringa leaves extract plus royal jelly (MLER) capsule supplementation on haematocrit level of anaemic pregnant woman in Takalar.

2. METHOD

This study was conducted in Puskesmas in North Polobangkeng sub-district, Takalar from July until September. Subjects of the study were 63 anaemic pregnant women who have signed approval sheet to be respondents. Samples were taken using simple random sampling based on inclusion criteria which were anaemic pregnant woman in 20-32 weeks of pregnancy, haemoglobin level (<11gr/dl), one foetus, not consuming multivitamin and mineral except Iron during experiment. Design of the study was True Experimental randomized double blind pretes-posttest controlled double blind design. Data was analysed using paired sample T-test and one way anova test.

Procedure Of Haematocrit Level Examnitaion

Study group were divided into 3 groups, 1 group got Moringa leaves extract (MLE) capsule, 1 group got Moringa leaves extract plus royal jelly (MLER) capsule, and 1 group got placebo. The blood samples of the subjects in intervention and control group had been taken in vena mediana cubiti as many as 3 cc before consuming the supplementation, the respondents then got explanation to consume 1 capsule a day with 1x500 mg doses for 60 days. After 60 days the blood samples were taken as post-test as many as 3 cc, and the blood samples were put into microsentrifugation tube contains EDTA as anticoagulant, then the blood samples were saved in in -700°C refrigerator before sent to laboratory to get haematocrit examination.

3. RESULT

Table. 1 showed most of anaemic pregnant women in intervention and control group were 20-35 years old. Most of them have high education, despite most of them were unemployed and have < Rp.3.1000.000 income. And most of the parity were primigravida.

	Group								
Characteristic	Moringa leaves extract plus royal jelly capsule (MLER)		Moringa leaves extract capsule (MLE)		Placebo (Control)		Total		p-Value
	n	%	n	%	n	%	n	%	
Age									
Low risk	16	33,3	6	33,3	16	33,3	48	100	1,000 ^a
High risk	5	33,3	5	33,3	5	33,3	15	100	
Education									
Low	7	3,3	6	28,6	8	8,1	21	100	0,807 ^a
High	14	33,3	15	35,7	13	31	42	100	
Job									
Unemployed	18	35,3	15	29,4	18	35,3	51	100	0,265 ^b
Employed	3	25	6	50	3	25	12	100	
Income									
< Rp.3.1000.000	19	33,9	17	30,4	20	35,7	56	100	0,158 ^b
\geq Rp.3.1000.000	2	28,6	4	57,1	1	14,3	7	100	
Parity									
Primigravida	10	30,3	11	33,3	12	36,4	33	100	$0,826^{a}$
Multigravida	11	36,7	10	33,3	9	30	30	100	

Table 1. Characteristic of Respondents in Intervention and Control Group

Chi-square^a, Mann-Whitney test^b

Based on Table. 2 average of haematocrit level in anaemic pregnant women in placebo group was lower than other groups. There was haematocrit level increase after intervention in MLER group, MLE group, placebo control group. The highest increase was in MLER with higher standard deviation than other groups. Statistic examination with significant value <0,05 showed that MLER capsule, MLE capsule, and placebo has effect on haematocrit level increase.

Advanced test was done using one way anova, average difference of haematocrit was twice higher in MLER group than MLE group and placebo group (control). Statistically p value in 3 groups was 0,000(<0,05), showed that there was difference in haematocrit level in MLER group, MLE group, and placebo group (control), interval trust value did not pass zero which meant there was difference of haematocrit level in MLER group and placebo group (control) clinically. But, the trust interval value in MLE passed zero. So, there was not difference of haematocrit level in MLE passed zero.

	Depend	lent Variable					
	Haematoo						
	Before	After	P-	Difference	CI	P-	
Independent	Average(±SD)	Average(±SD)	value		95%	value	
Variable							
Moringa Leaves	30,12(±1,88)	34,66(±2,88)	0,000 ^a	4,53	3,36-		
Extract Plus				$(\pm 2,58)$	5,71		
Royal Jelly							
(MLER)						0,000 ^b	
Moringa Leaves	31,21(±1,28)	33,38(±2,16)	0,000 ^a	2,17	-3,01-		
Extract (MLE)				(±1,85)	1,32		
Placebo	31,51(±1,83)	32,62(±2,40)	0,046 ^a	1,11	0,02-		
(control)				(±2,40)	2,20		

Table. 2 Effect of Before and After Moringa Leaves Extract Plus Royal Jelly Capsule Supplementation, Moringa Leaves Extract capsule, and Iron Supplementation (control) on Haematocrit Level

Paired sample T-test^a, One Way Anova^b

Table. 3 was advanced analysis using post-hoc Bonferroni to know which intervention group which showed significant difference clinically. The table showed all of the groups have significant difference clinically and statistically. So, it can be concluded that MLER, MLE, and placebo (control) have effect on haematocrit level increase.

Table. 3 Analysis Post-Hoc Difference of Haematocrit Level in Moringa Leaves Extract Pl	'lus
Royal Jelly Group, Moringa Leaves Extract Group, and Iron Supplementation Group	

Group	Average Difference	CI95%	P Value
Moringa Leaves Extract Plus Royal Jelly vs Moringa Leaves Extract	6,71(±3,81)	4,96-8,46	0,000
Moringa Leaves Extract Plus Royal Jelly vs Placebo (control)	3,42(±3,82)	1,67-5,17	0,000
Moringa Leaves Extract vs Placebo (control)	3,28(±2,98)	-5,04-1,54	0,000

Post-hoc Bonferroni

4. **DISCUSSION**

Result of the study of the effect and difference of MLER capsule, MLE capsule, and placebo on haematocrit level in table 2 and 3 showed that the highest increase of haematocrit level in in anaemic pregnant women happened in pregnant women who consumed Moringa leaves extract plus royal jelly supplementation. This is equal to study by Lutfiyah Fifi (2011) that showed that Moringa leaves powder with 1, 2, and 3 doses has effect on representation of sample's haematology. Nutrient composition of Moringa leaves powder from NTB has been examined and showed that the protein level was equal to Moringa leaves powder in Africa countries. Beside protein, component which has role in increasing Haemoglobin, erythrocyte, albumin, iron, and haematocrit¹². Study by Adedapo (2009) with 54 male mice which were divided into 1 distillated water group, and 3aquesous Moringa leaves extract oral for 21 days intervention groups. There was significant increase of heametocrit level ($45,3\pm1,6\%$) in 400 mg/kg group, but there was not significant haemoglobin change (14,0 \pm 1,7 g%) and amount of erythrocyte (7,4 \pm 0,1 million/mm3). Group with 800 mg/kg experienced significant decrease of haematocrit, haemoglobin, and erythrocyte counts. While in 1600 mg/kg doses, haematocrit decreased significantly but followed by insignificant haemoglobin change and amount of erythrocyte ¹¹.

Haematocrit is percentage of whole blood volume which consists of erythrocyte. Anaemia in pregnant woman is marked by haematocrit level decrease because of blood volume increase and unbalanced blood cell production in pregnancy, so blood volume increase for about 42% while erythrocyte increase for about one third of it so physiology haemodilution happens which causes haemoglobin and haematocrit level decrease. Study by Samuel et.al (2015) showed that Moringa leaves was beneficial for anaemic patient whether in low or relative high dose. There was significant difference of erythrocyte and leucocyte amount with Moringa leaves supplementation, it showed that it was not only good for food supplementation but also as medicine especially for anaemic patient¹³.

After given supplementation in the morning for 60 days, blood samples were taken to measure haematocrit level using Heamatologi Analyze and compared haematocrit level before and after intervention, it showed that haematocrit level in anaemic pregnant women increased. It was because pregnant women who consumed Moringa leaves extract plus royal jelly supplementation were more relaxed, had better appetite and sleep pattern than before consuming the supplementation. There was statistically significant difference and effect on body weight and haematocrit level of pregnant women who consumed MLE or MLER. It could be caused by not enough amounts of samples. But, consuming MLE or MLER both could increase body weight and haematocrit level.

Good nutrient intake during pregnancy is important because consuming enough macronutrient and micronutrient is beneficial to fulfil nutrient needs during pregnancy. Nutrient status of pregnant woman is influenced by intake quantity of energy or calorie, protein, iron, carbohydrate, vitamin A, folic acid, calcium, iodine, and other nutrients. In all of country, pregnant woman and children under 5 years old is on the highest risk of micronutrient deficiency. Deficiency of iron, iodine, folic acid, vitamin A, and zinc is the widest micronutrient deficiency and common contributor of bad growth, intellectual interruption, perinatal complication, and increase of morbidity and mortality⁶.

5. CONCLUTION

Moringa leaves extract plus royal jelly capsule supplementation, Moringa leaves extract capsule and palcebo influenced significantly in increasing haematocrit level, so this supplementation can be given to anaemic pregnant woman.

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