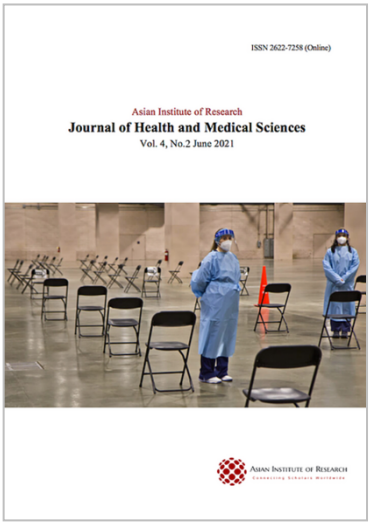

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Protein and Iron Bioavailability, Perception, Menstrual Cycle as Adolescent Girls' Anemia Factors

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Abstract
Adolescent girls are one of the groups of people who is prone to iron nutrient deficiency. Iron is required as a substitute for iron lost due to the menstrual cycle. This research aims to determine the trigger factors of anemia in adolescent girls who become participants of the prevention and control program of anemia. This type of research is an observational research with cross sectional design and using statistical test of chelstle method of Mantel Haentzel and OR value for its meaning. The results shows there are four significant triggers of anemia that is perception of adolescent about nutrition (OR = 2,24; 95% CI = 1,05 - 4,76), adherence to TTD (OR = 2,49; 95% CI = 1.11 - 5.58), protein consumption levels (OR = 3.27, 95% CI = 1.57 - 6.84), iron intake (OR = 2.81; 95% CI = 1.30 - 6.05), and duration of menstrual bleeding (OR = 8.08; 95% CI = 1.05 - 61.89). The distribution of blood booster tablets or *tablet tambah darah* (TTD) needs to be intensified again, accompanied by an emphasis on the benefits of TTD tablets for young women, and to continue to consume independently when the distribution of TTD is terminated. In conclusion, adolescent girls are prevalent to iron nutrient due to menstrual cycle. Therefore, nutrition counseling should also be given besides consuming fresh foods rich in protein and iron as well as vegetables and fruits, because both foods contain vitamin C which greatly helps the absorption of iron in the body.

Keywords: Adolescent Girls, Anemia, Nutrition, Risk Factors

1. Introduction

Adolescent girls are group of people who is to iron deficiency because they are in a peak growth. The need for iron in a high amount is needed, especially for basal metabolism. The rapid growth in this phase has the consequence in the increase of the need for nutrients. In this phase, it is often found that adolescent girls prefer strict diets that tend to deviate. More than 50.0% of the incidence of anemia is caused by iron deficiency. Low iron consumption is considered as major cause of suffering from anemia (Jamnok et al., 2020; Miller, 2013). There is a relationship between protein intake and anemia (Gasong et al., 2019). Low protein intake has the risk 3.48 times greater for suffering from anemia. Another factor that also contributes to anemia is the low absorption

of iron from food. The absorption of iron requires an acidic environment with vitamin C as the reductors. Besides, Vitamin A is required in hematopoiesis.

The balance of iron in the body is important for metabolism. The amount of daily turnover of iron is 35 mg, and not all of it comes from food. Around 34 mg is obtained from the destruction of old red blood cells that is filtered by the body to utilize again in form of new red blood cells. The impact of anemia on adolescent girls are stunting, susceptibility to infection, decreased immune system, decreased fitness or freshness, decreased enthusiasm for learning and achievement (Chaparro & Suchdev, 2019; Wayan et al., 2020).

The modern lifestyle of adolescents in today's time is commonly in line with society, tends to be with the pattern of low iron bioavailability. They consume more rice, tubers, nuts, and eat less meat, chicken, or fish, which are low in vitamin C. The regular lifestyle that is visible is like food restrictions up to uncontrolled eating habits. Implementation of dietary restrictions arise because of concerns with obesity. On the other hand, food supplements are now an alternative for adolescent girls to consume. Nutritional status, described as body mass index (BMI), is known to have a positive correlation with hemoglobin concentration. Low BMI or underweight body has 1.5 times the risk of suffering from anemia. Clean and healthy living behavior or *Perilaku Hidup Bersih dan Sehat* (PHBS) is one of the leading programs that has been disseminated for a long time. The main goal is to form healthy behavior to avoid infectious diseases such as worm infestation. Approximately 50.0% of the rural population, who suffer from anemia caused by iron deficiency and 40% of iron deficiency anemia accompanied by hookworm infection (Rodríguez-Guardado et al., 2013; Ngui et al., 2012). The data of anemia and its risk factors for adolescent girls in several areas are relatively small, not comparable to the very high number of adolescent girls (18.4%) and keep increasing. The prevalence of anemia in adolescent girls is 22.3% (Dinas Kesehatan Provinsi Bali, 2014).

2. Method

This observational study with a cross sectional design was conducted from May to August 2017. The research was conducted in Badung Regency, Bali with the target population was female students who participated in the anemia prevention and control program. The number of samples was 200 people who were selected simply and randomly.

The data collection includes the incidence of anemia. In collecting the data, this research used an instrument in the form of the SQ-FFQ form (Semi Quantitative Food Frequency Questioner) to assess the bioavailability of nutrient consumption; a questionnaire to collect data on knowledge, perceptions, hygiene-healthy behavior, menstrual cycle, information on worms infestation; and digital laboratory tools for the determination of hemoglobin levels. The weight measuring instrument used is a digital weighing scale Fesco, while for height, we used Microtoise One med.

The knowledge and perception data were collected through interviews by filling in answers to the available questionnaires or lists of questions. Also, the data on iron bioavability and food supplements were collected by doing interviews using the SQ-FFQ form. Meanwhile, the data on PHBS were collected by interview covering habits of maintaining body hygiene including nails, hair and teeth. The worm infestation data were collected by doing interviews regarding the habit of consuming deworming drugs in a certain period (frequency, and type or brand of the drug consumed). Next, the data on Body Mass Index were collected through weighing and measuring body height, which were compared with the ideal standards. The data of menstrual cycle is done by interviewing the participants using the menstrual description list form. The last but not least, the hemoglobin data were collected by doing laboratory examination methods using the Esaytouch device.

Since all the following variables was converted into dichotomous variables (there are only two categories), Chi-Square statistical analysis is used to analyze the relationship between various independent variables and the dependent variable on anemia (Mantel Haentzel).

3. Results

Although it is not explicitly included in high school curriculum, understanding about nutrition has become general knowledge which can be obtained from various other learning sources (Vlieger et al., 2020; Craigieet al.l, 2011). The sample's knowledge and perceptions of nutrition are considerably various. The samples having high knowledge (48.8%) are almost the same as the low ones (51.2%), so are their perceptions of nutrition. The number of samples that tend to have positive perception (48.3%) is almost the same as those with negative perception (51.7%).

Not all samples have demonstrated clean and healthy living behavior (PHBS). A small percentage of the sample (25.4%) applies regular hygiene and sanitation. Besides, those who are accustomed to consuming supplements are much less, that is only 7.5% as well as consumption of deworming drugs with only 14.9% of the sample and even then it was not done regularly every 6 months. On the other hand, most of the samples (79. 6%) had taken blood booster tablets or *tablet tambah darah* (TTD) as shown in Table 1. This is because the program for giving such tablets to adolescent girls has been conducted through the School Health Unit or *Usaha Kesehatan Sekolah* (UKS).

Table 1: Sample distribution for clean and healthy living behaviour

Variable	**Category**	**Observation Results**	
		F	**%**
Sanitation and Hygiene Behavior	Unorganized	150	74.6
	Well-organized	51	25.4
	Total	**201**	**100.0**
Supplement Consumption	Yes	15	7.5
	No	186	92.5
	Total	**201**	**100.0**
Blood Booster Consumption	Yes	160	79.6
	No	41	20.4
	Total	**201**	**100.0**
Deworming Drug Consumption	Yes	30	14.9
	No	171	85.1
	Total	**201**	**100.0**

The implementation of PHBS will have an impact on the immune system to resist the diseases. Table 2 shows the results of observations found most samples suffered from diarrhea during the past month and it was only 9% of the sample.

Table 2: Sample Distribution of Disease

Experiencing Diarrhea for the past month	**Observation Result**	
	f	**%**
Yes	18	9.0
No	183	91.0
Total	**201**	**100.0**

In this study, the menstrual cycle of the sample was also observed because it seemed to play a role as a trigger for anemia to adolescent girls. Table 3 shows half of the samples (67.3%) experienced menstruation regularly once a month and most of them (84.6%) had period length for more than 3 days. Also, the level of consumption of sample nutrients still does not meet the requirements for nutritional balance.

Table 1: Sample Distribution of Menstrual Cycle

Variable	Category	Observation Result	
		F	%
Menstrual Period Frequency	Regular	109	67.3
	Irregular	53	32.7
	Total	**162**	**100.0**
Length of period	≤ 3 days	25	15.4
	> 3 days	134	84.6
	Total	**162**	**100.0**

Nearly all samples (95%) consumed proper carbohydrates even more than enough. For protein intake, only about 70.6% of the sample consumes sufficient amounts. Even specifically for the consumption of iron as the main factor for the production of red blood cells, only 50.2% of the sample has sufficient amount of iron intake. Moreover, Vitamin C, a nutrient that helps the absorption of iron, has almost reached the level of consumption according to balanced nutrition guidelines. Table 4 shows the data that only 16.4% of the sample consumed less Vitamin C than the recommended amount. The nutritional condition of the sample is quite good. Most of the samples have normal nutritional status.

Table 2: Sample Distribution of Nutrient Consumption Levels

Nutrients	Category	Observation Result	
		F	%
Energy	Deficient	37	18,4
	Sufficient	164	81.6
	Total	**201**	**100.0**
Carbohydrate	Deficient	10	5.0
	Sufficient	191	95.0
	Total	**201**	**100.0**
Fat	Deficient	86	42.8
	Sufficient	115	57.2
	Total	**201**	**100.0**
Protein	Deficient	59	29.4
	Sufficient	142	70.6
	Total	**201**	**100.0**
Iron	Deficient	101	50.2
	Sufficient	100	49.8
	Total	**201**	**100.0**
Vitamin C	Deficient	33	16.4
	Sufficient	168	83.6
	Total	**201**	**100.0**

A small proportion of the sample were skinny (2.5%), as well as the fatty ones, only 10% of the sample as shown in Table 5. According to the results of the hemoglobin examination that has been carried out, the percentage of anemia problems is classified as low, as it is only 37 samples (18.4%) suffer from anemia. Table 6 shows that those with anemia, 56.8% of the sample had low nutritional knowledge, while 50% of the group without anemia also had low nutritional knowledge. Samples with low nutritional knowledge were more likely to be found in those with anemia. Statistically, this trend proved to be insignificant with a value of χ^2=0.55 (p>0,05).

Table 3: Distribution of Nutritional Status Sample According to BMI/Age Index

Category	**Observation Result**	
	F	**%**
Fatty	20	10,0
Normal	176	87.6
Skinny	5	2.5
Total	**201**	**100.0**

Based on perceptions about nutrition, the tendency for differences in the incidence of anemia between groups is clearer. In the group who had anemia, 67.6% of the sample had negative perceptions, while in the group without anemia, only 48.2% had negative perceptions shown in Table 6. Therefore, the results of these observations found that the sample who had negative perceptions about nutrition was more likely to experience anemia than those who did not. From the results of statistical analysis, there was a significant difference with the value χ^2=4,55 (p≤0,05). There are differences in the incidence of anemia related to the sample's perception about nutrition.

Table 4: Sample Distribution of anemia status according to the level

Variable	Category	Sample of Anemia Status				Total	
		Anemia		No			
		F	%	F	%	f	%
Level of knowledge of nutrition	Low	21	56.8	82	50.0	103	51.2
	High	16	43.2	82	50.0	98	48.8
Total		37	100.0	164	100.0	201	100.0
Trends in perception of nutrition	Negative	25	67.6	79	48.2	104	51.7
	Positive	12	32.4	85	51.8	97	48.3
Total		37	100.0	164	100.0	201	100.0

According to the data about practice of sanitation hygiene, 86.5% of those in the group did not practice regular hygiene and sanitation, and those in the group that did not suffer from anemia, 72% did not practice regular hygiene. This implies that the sample who did not practice regular hygiene and sanitation was more likely to be found in those who had anemia than those who did not. However, statistically the trend of this difference was not significant with a value of χ^2=3.37 (P>0.05).

A contrast result is shown on the habit of consuming supplements. The samples who had anemia, 89.2% did not regularly take supplements, whereas in the samples who did not have anemia, 93.3% did not usually take supplements either. Thus, the data shows that the samples who do not usually take supplements are more likely to not suffer from anemia than those who do. However, it statistically does not show a significant difference with a value of χ^2=0.74 (p>0.05).

This is also found in the consumption of deworming medicine. In the sample group who had anemia, 83.8% of them did not take deworming medicine, whereas in the sample group that did not have anemia there were 85.4% of them did not take deworming medicine. Hence, the data shows that the samples that did not take deworming medicine were more likely did not suffer from anemia than those who did. However, same with the consumption of supplements, this difference was insignificant with a value of χ^2=0.06 (p>0.05).

A different fact is found in the consumption of blood or Iron booster tablets (TTD) as one of the Health Office programs to prevent anemia from adolescent girls. The distribution of iron tablets for adolescent girls has been declared as one of the School Health Unit (UKS) programs so that almost the majority of the sample already consumed such tablets. In the sample group who had anemia, 32.4% of the sample did not consume iron tablets and those in the non-anemia group only 15.9% who did not consume it. Therefore, the samples that did not consume iron tablets were more likely to get anemia than those who did not. This difference was statistically

significant with a value of χ^2=5.14 (p≤0.05). There are differences in the incidence of anemia according to the habit of consuming iron tablets.

Besides, the data on the frequency of menstruation shows such a contrary results. Regarding the irregular menstrual frequency, there was 21.2% of the samples in the group of anemia, and 35.7% was in the group of those who did not have anemia. This means that there were more samples who experienced irregular menstruation in the group that did not experience anemia than the group who suffer from such disease. However, statistics shows that there was no significant difference in the incidence of anemia according to the frequency of menstruation with a value of χ^2=2.49 (p>0.05).

Meanwhile, according to the length of bleeding during menstruation, there were 97% samples who had anemia experienced bleeding during menstruation for more than 3 days and those in the group who did not get anemia, only 79.8% of the sample experienced bleeding during menstruation for more than 3 days. The sample who experienced bleeding during menstruation for more than 3 days was found more in the group that had anemia than the group that did not. The statistical analysis indicates that the difference is significant with a value of χ^2=5.55 (p≤0.05). Thus, there is a significant difference in the incidence of anemia related to the length of bleeding during menstruation.

In the group with anemia, 16.2% of the sample had less energy consumption, while in the non-anemia group, the sample with less energy consumption was higher (18.9%). Samples with less energy consumption were more likely to not experience anemia than those who did. However, this difference was statistically insignificant with a value of χ^2=0.15 (p>0.05).

If it is observed from the protein level, 51.4% of the sample in anemia group were less in protein consumption level, while the sample group that did not experience anemia there were only 24.4% of the sample whose protein consumption level was less. Thus, samples with less protein consumption levels were more likely to be found in the sample group who had anemia than those who did not. Based on statistical analysis, a significant difference is obtained with the value χ^2=10.58 (p≤0.05). There is a significant difference in the incidence of anemia based on the level of protein consumption.

According to the data of fat consumption level, the opposite result is found. 37.8% of the sample in the group suffering from anemia had less fat consumption, while the non-anemia group had more samples that is 43,9% as shown in Table 7. There are more sample in the group who do not have anemia whose level fat consumption is low or deficient than those in the anemia group. However, this difference was not statistically significant with a value of χ^2=0.45 (p>0.05).

Table 5: Distribution of anemia status according to level consumption of sample nutrient

Nutrient	Category	Sample of Anemia Status				Total	
		Anemia		No			
		f	%	F	%	f	%
Energy	Deficient	6	16.2	31	18.9	37	18.4
	Sufficient	31	83.8	133	81.1	164	81.6
Total		37	100.0	164	100.0	201	100.0
Protein	Deficient	19	51.4	40	24.4	59	29.4
	Sufficient	18	48.6	124	75.6	142	70.6
Total		37	100.0	164	100.0	201	100.0
Fat	Deficient	14	37.8	72	43.9	86	42.8
	Sufficient	23	62.2	92	56.1	115	57.2
Total		37	100.0	164	100.0	201	100.0
Carbohydrate	Deficient	1	2.7	9	5.5	10	5.0
	Sufficient	36	97.3	155	94.5	191	95.0
Total		37	100.0	164	100.0	201	100.0
Iron	Deficient	26	70.3	75	45.7	101	50.2

	Sufficient	11	29.7	89	54.3	100	49.8
Total		37	100.0	164	100.0	201	100.0
Vitamin C	Deficient	9	24.3	24	14.6	33	16.4
	Sufficient	28	75.7	140	85.4	168	83.6
Total		37	100.0	164	100.0	201	100.0

The same result is also presented in level of carbohydrate intake. In the group having anemia, 2.7% of the sample had a low level of carbohydrate consumption, while in the non-anemia group, there are 5.5% sample whose level of carbohydrate consumption was low. Thus, there are more samples whose carbohydrate consumption is deficient in the group who did not have anemia than those who did. However, this difference was not statistically significant with a value of χ^2=0.50 (p>0.05).

Contrary to the level of consumption of other nutrients, there is a clear proclivity in the level of iron intake. In the group that had anemia, there were 70.3% of the sample whose iron intake was insufficient, and in the sample group that did not have anemia there were only 45.7% whose level of iron intake was insufficient. Thus, the samples with insufficient iron consumption levels were more likely to be found in the sample group who had anemia than those who did not. Therefore, the statistical analysis shows a significant difference with a value of χ^2=7.27 (p≤0.05). This implies that there is indeed a significant difference in the incidence of anemia when viewed based on the level of iron consumption.

And the last but not least, there was no significant difference at the level of vitamin C consumption. In the group with anemia, 24.3% of the sample had a low level of vitamin C consumption, while in the non-anemia group, the sample whose consumption level was insufficient was only 14.6%. However, this difference was not statistically significant with a value of χ^2=2.07 (p>0.05).

Based on the nutritional status there is a reciprocal tendency so that the correlation between anemia status and nutritional status of the sample becomes difficult to predict. For the sample with obesity, those who did not experience anemia (11.6%) were higher than those who had anemia (2.7%). However, in samples with normal nutritional status, more people had anemia (94.6%) than those who did not (86%) as shown in Table 8. Based on statistical analysis, there was no difference in the incidence of anemia related to the nutritional status of the sample with a value of χ^2=2.66 (p>0.05).

Table 6: Distribution of Anemia Status Based on Sample of Nutritional Status

Nutritional Status	**Sample of Anemia Status**				**Total**	
	Anemia		**No**			
	F	**%**	**f**	**%**	**f**	**%**
Fatty	1	2.7	19	11.6	20	10.0
Normal	35	94.6	141	86.0	176	87.6
Skinny	1	2.7	4	2.4	5	2.5
Total	37	100.0	164	100.0	201	100.0

4. Discussion

There are five factors that trigger significant incidence of anemia, namely perceptions of nutrition, consumption of blood booster tablets, level of protein and iron intake, and bleeding during menstruation (Gautam et al., 2019; Triharini et al., 2018; Thomson et al., 2012; Abbaspour et al., 2014; Sumarlan et al., 2018).

Based on the risk factor analysis, the perception of nutrition has a value of OR = 2.24 with 95% CI = 1.05 - 4.76. This means that significantly samples with negative perceptions of nutrition have 2.24 times greater risk of anemia than those with positive perceptions. Misconceptions in interpreting the appearance of the body such as get fatty, as well as other negative perceptions have the potential for adolescent girls to experience eating disorders or take a strict diet to achieve body goals. Also, misperceptions about body appearance which leads to eating disorders often cause serious health problems such as experiencing malnutrition due to strict diets,

depression, feeling inferior to the environment, mental health problems and even suicide. It is important to note various factors which can affect such thing to happen. Parents' comments regarding the child's appearance, the role of the media, psychological, social, and cultural values in society are some of the factors that need to be considered. All efforts should be done by involving various parties, both private and government as well as parents to provide correct information and as an effort to promote health for children and adolescents to prevent misunderstandings about body appearance and eating disorders (Golden et al., 2021; Leme et al., 2020).

Also the risk factor analysis shows that adherence with iron or blood booster tablets (TTD) intake has a value of OR = 2.49 with 95% CI = 1.11 - 5.58. This means that those who did not adhere with iron tablets intake had 2.49 times greater risk of anemia than those who did. In the guidebook for the prevention and control of anemia in young and eligible women, it is necessary to obtain iron supplementation when food consumption is not sufficient enough to provide the needs of iron. The distribution of iron or blood booster supplement for a certain period of time aims to increase hemoglobin levels rapidly, and it is necessary to continue to increase iron stores in the body. Supplementation of Blood Booster Tablets (TTD) for young and eligible women is one of the efforts of the Indonesian government to meet iron intake. Giving iron tablet in the right dose can prevent anemia and increase iron reserves in the body (Fishman et al., 2000; Ministry of Health of Indonesia, 2016; Finkelstein et al., 2018; Stoffel et al., 2020).

From the risk factor analysis, level of protein consumption has a value of OR = 3.27 with 95% CI = 1.57 - 6.84. This means that significantly the sample whose protein consumption is less than the recommended nutritional adequacy rate has 3.27 times greater risk of anemia than the sample whose protein consumption meets the recommended rate. The percentage of anemia incidence is higher in the elderly who have low protein consumption level than those who consume enough protein (Bianchi, 2015). Low protein intake will interfere with the transport, production, and storage of iron. There are three types of proteins interrelated in the process of transporting and storing iron in the body, namely transferrin, the transferrin receptor 1 (TfR1) and ferritin. Transferrin transports iron to tissues that have transferrin receptors, especially erythroblasts in the bone marrow for the process of hemoglobin formation, so that lack of protein consumption does have a direct impact on the incidence of anemia as a result of failure of hemoglobin formation in the spinal cord (Alamsyah & Andrias, 2016).

The risk factor analysis also notes that the level of iron consumption has an OR = 2.81 with a 95% CI = 1.30 - 6.05. This means that significantly the sample whose iron consumption is less than the recommended nutritional adequacy rate has 2.81 times greater risk of anemia than those whose iron consumption fulfills the recommended rate. This is in line with research conducted by (Astuti, 2010) by obtaining a strong correlation (r = 0.675) between iron intake and hemoglobin levels in 122 children aged 7 to 15 years old in Kulon Progo, Yogyakarta. These synergistic results actually reinforce the fact that low consumption of iron-rich foods can disrupt the formation of red blood cells so that the hemoglobin level in the body decreases which can eventually lead to anemia.

Additionally, the risk factor analysis also shows the length of bleeding during menstruation has a value of OR = 8.08 with 95% CI = 1.05 - 61.89. This means that samples whose bleeding during menstruation were more than 3 days have 8.08 times greater risk of anemia than that of samples whose bleeding were less than 3 days. Similar results were also obtained in the research of (Febrianti et al., 2013) by taking a sample of 250 female students of Madrasah Aliyah Negeri 2 Bogor. The conducted observation found that there was a significant relationship between the length of menstruation and the incidence of anemia in adolescent girls (p value = 0.028). The incidence of anemia in this study was significantly related to length of period. Forty percent (40%) of female students in this study experienced menstruation for more than 7 days. Menstruation more than 7 days is one of the symptoms of menorraghia. Menorrhagia is the medical term for periods with bleeding that is more than normal or longer than normal. Such thing can happen due to hormonal imbalance, ovarian dysfunction, uterine fibroids, polyps in the uterine wall, adenomyosis, intrauterine devices, pregnancy complications, cancer, genetic disorders, consumption of certain drugs, or other medical conditions.

Conclusion

The prevalence of anemia on adolescent girls participating in anemia prevention and control programs is 18.4%. Among the six types of nutrients analyzed for their correlation to the incidence of anemia in adolescent girls, namely energy, carbohydrates, protein, fat, iron, and vitamin C, only the level of protein and iron intake shows a statistically significant difference. Hence, there is indeed a significant difference in the incidence of anemia according to the bioavailability of iron consumption. According to the nutritional status, there is an opposite tendency so that the relationship between anemia status and nutritional status of the sample becomes difficult to predict. Based on statistical analysis, there is also no difference in the incidence of anemia related to the nutritional status. Different things happen from the length of bleeding during menstruation. It is proven that more samples who experienced menstrual bleeding for more than 3 days were found in the anemia group compared to the group that did not suffer from it. Based on statistical analysis, this difference is significant so that that there is indeed a difference in the incidence of anemia based on the length of bleeding during menstruation.

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