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Local Food-Based Intervention on Nile Tilapia (Oreochromis niloticus) and Banana Blossom (Musa balbisiana Colla) On Lactoferrin Levels In Breastfeeding Mothers

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Abstract

Lactoferrin is a bioactive glycoprotein in breast milk (ASI) that plays a vital role in the infant's immune system. Lactoferrin levels in breast milk can be influenced by the mother's nutritional status and dietary intake. Consuming local foods rich in protein and containing galactagogues has the potential to improve breast milk quality, including lactoferrin. This study aimed to analyze the effect of a locally processed food intervention on lactoferrin levels in breastfeeding mothers. The study was conducted from April to July 2025 in Tasikmalaya Regency, West Java, Indonesia. This study used a quasi-experimental pre-test–post-test control group design with 80 breastfeeding mothers with 14-day-old infants recruited from six community health centers. The intervention group (n=40) received 200 g of tilapia (Oreochromis niloticus) and 200 g of banana blossom (Musa balbisiana Colla) per day for 14 days, along with breastfeeding promotion. The control group (n=40) received only breastfeeding promotion. Lactoferrin levels were analyzed using the ELISA method, and data were analyzed using paired and independent t-tests, Wilcoxon, Mann–Whitney, and the General Linear Model. The results showed that breastfeeding mothers who received the intervention experienced a greater increase in lactoferrin levels compared to the control group. Changes in lactoferrin levels (Δ lactoferrin) were significantly different between the two groups ($p = 0.032$), and the General Linear Model analysis showed that the intervention had an effect on these changes ($p = 0.050$). Giving tilapia and banana blossom for 14 days increased lactoferrin levels in breastfeeding mothers.

Introduction

Infant mortality remains a public health problem in various developing countries, including Indonesia. Data shows that the infant mortality rate in Indonesia remains relatively high, at around 16.85 per 1,000 live births, ranking third highest in Southeast Asia (Nurfirdaus & Bassey, 2021; Rumiati & Adisasmita, 2021). West Java Province is one of the regions with a significant contribution to infant mortality in 2023, and Tasikmalaya Regency is among those with a high infant mortality rate. Efforts to reduce infant mortality require a variety of approaches, one of which is improving the quality of breastfeeding (Brockway, 2024; Yasir, 2019). Breast milk is the best source of nutrition for babies because it provides a variety of macronutrients and micronutrients needed for growth and development (Indrayani *et al.*, 2020). Furthermore, breast milk contains various bioactive components that play a vital role in the baby's immune system, one of which is lactoferrin (Lawrence, 2022). Lactoferrin is a glycoprotein that plays an important role in the immune system, iron transport, and protection against infection. It is known to have antimicrobial, anti-inflammatory, and immunomodulatory activities that support

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the development of the baby's immune system (Legrand *et al*., 2008).

Lactoferrin levels in breast milk are not constant and are influenced by various factors. Concentrations are highest in colostrum (>5 g/L), then decrease in mature milk (2–3 g/L), and can increase again during prolonged lactation (Czosnykowska-Łukacka *et al*., 2019; Villavicencio *et al*., 2017). Apart from the lactation stage, other factors such as the mother's psychological condition, prematurity of the baby, environmental exposure, frequency of breastfeeding, and parity also contribute to variations in lactoferrin concentration (Villavicencio *et al*., 2017; Wu *et al*., 2024; Y. Zhou *et al*., 2025). In addition, the intake of nutrients such as protein (M. Zhou *et al*., 2025), lipid(Wu *et al*., 2024), iron (Ruiz-Rico *et al*., 2025), zinc (Dumrongwongsiri *et al*., 2021), calcium (Dumrongwongsiri *et al*., 2021), selenium, and vitamin B-12 (Gibson *et al*., 2020) have also been reported to be associated with lactoferrin levels in breast milk.

Maternal dietary intake is a modifiable factor that plays a role in influencing the bioactive composition of breast milk, including lactoferrin levels. Nutrients such as protein and iron are known to play a role in the synthesis and biological activity of lactoferrin. Consistent with this, previous research has shown that protein and energy consumption during lactation is positively correlated with lactoferrin levels in breast milk. In fact, every 1-gram increase in egg consumption has been reported to increase breast milk lactoferrin levels by 0.3 μg/mL, demonstrating the importance of adequate nutritional intake in supporting the immunological quality of breast milk (Liu *et al*., 2022). Therefore, the use of local foods rich in protein and iron, such as tilapia (*Oreochromis niloticus*) and banana blossom (*Musa balbisiana* Colla), has the potential to support increased lactoferrin levels in breastfeeding mothers.

Local food potential can be utilized as a nutritional source to support breast milk quality in breastfeeding mothers. Tasikmalaya Regency has abundant local food sources from the agricultural and fisheries sectors, including banana blossoms (*Musa balbisiana* Colla) and tilapia (*Oreochromis niloticus*). Banana blossoms are known to contain iron and various bioactive compounds, while tilapia is a good source of animal protein. However, research specifically evaluating local food interventions on lactoferrin levels in breastfeeding mothers is still very limited, especially in the context of local food utilization at the community level. Therefore, this study aims to evaluate the effect of tilapia and banana blossom interventions on lactoferrin levels in breastfeeding mothers.

Method

This quasi-experimental study employed a pre-test–post-test control group design and was conducted from April to July 2025 in Tasikmalaya Regency, West Java Province, Indonesia. Six Community Health Centers (Puskesmas) were selected as study sites and randomly assigned to intervention and control groups, with three Puskesmas in each group. The study population consisted of breastfeeding mothers residing in Tasikmalaya Regency. Eligible participants were mothers on the 14th day postpartum who were exclusively breastfeeding, had a normal body mass index (BMI), did not experience postpartum infections or complications, and agreed to participate in the study. Respondents who dropped out or did not complete the study procedures were excluded. The sample size was calculated using the mean difference formula for two independent numerical groups based on parameters from previous studies, with a significance level of 5% and a statistical power of 90%. The minimum required sample size was 36 participants per group. To account for possible loss to follow-up, four additional participants were included in each group, resulting in a total sample of 80 breastfeeding mothers, comprising 40 participants in the intervention group and 40 participants in the control group. Participants were recruited consecutively from the selected Puskesmas until the required sample size was achieved.

The intervention group received 200 g of processed Nile tilapia (*Oreochromis niloticus*) and 200 g of processed banana blossom (*Musa balbisiana Colla*) once daily for 14 consecutive days, in addition to breastfeeding promotion delivered four times during the study period. The control group received breastfeeding promotion only. Fresh tilapia and banana blossoms were obtained from local markets in Tasikmalaya Regency and processed using standardized recipes. Banana blossoms were cleaned, sliced, boiled, and prepared into several local dishes, including pepes banana blossom, dendeng banana blossom, shredded banana blossom, and cobek banana blossom. Tilapia was cleaned, marinated, and prepared as steamed, fried, grilled, or yellow-spiced dishes. Rotating menus were provided throughout the intervention period to improve acceptability and reduce participant fatigue. Compliance with food consumption was monitored through direct observation and daily consumption records. Baseline characteristics collected included maternal age, parity, postpartum depression screening score, breastfeeding frequency, and breastfeeding duration. Dietary intake was assessed using a 2 × 24-hour food recall to evaluate the adequacy of energy, protein, fat, vitamin A, vitamin B12, iron, calcium, and zinc intake, expressed as percentages of recommended dietary requirements. Measurements were performed twice, namely before the intervention on day 13 postpartum and after the intervention on day 28 postpartum.

The primary outcome variable was maternal serum lactoferrin level. Venous blood samples (3 mL) were collected by trained laboratory personnel at baseline and post-intervention using serum separator tubes (SST). Samples were centrifuged at 3000 rpm for 10–15 minutes to obtain serum, transferred into sterile microtubes, and stored at −80°C. The serum samples were temporarily stored at KHZ Tasikmalaya Hospital and subsequently transported in a cooled container to the GAKI Laboratory, Diponegoro University, for analysis. Lactoferrin concentrations were measured using the Enzyme-Linked Immunosorbent Assay (ELISA) method according to the manufacturer's protocol and expressed in μg/mL. Data were analyzed using paired t-tests, independent t-tests, Wilcoxon tests, Mann–Whitney tests, and the General Linear Model. Potential confounding variables, including maternal age, parity, breastfeeding practices, and nutrient intake, were considered in the analysis. Ethical approval was obtained from the Health Research Ethics Committee of the Faculty of Public Health, Diponegoro University (No. 123/EA/KEPK-FKM/2025), valid from March 27, 2025, to March 27, 2026.

Result and Discussion

A total of 80 breastfeeding mothers were recruited at the beginning of the study, and all respondents completed the study until the final stage, resulting in all data from 80 respondents being analyzed. The baseline characteristics of the study participants included maternal age, parity, depression screening results, and breastfeeding frequency. In general, the background characteristics of the respondents were relatively homogeneous across the study groups, as presented in Table 1.

TABLE 1. Baseline characteristics of breastfeeding mothers by study group

Characteristics	**Intervention (n=40)**		**Control (n=40)**		**p value**
	x̄ (±SD)	**Med(Min-Max)**	**x̄ (±SD)**	**Med (Min-Max)**	
Maternal age (years)		27.70(21-42)		28.00(19-40)	0.650
Parity (number)		2.00(1-6)		2.00(1-4)	0.649
Postpartum depression screening (score)					
Before	9.05(±3.15)		9.10(3.38)		
After	8.55(±3.51)		8.28(3.68)		
Breastfeeding frequency (times/day)					

Before	12.00(7-18)	12.00(9-33)	0.497
After	12.00(9-20)	12.00 (10-34)	0.444
Breastfeeding Duration (minutes)			
Before	20.00(10.00-38.00)	21.00(10.00-34.00)	0.444
After	20.00(6.00-34.00)	20.00(10.00-33.00)	0.400

*Values are presented as mean ± SD or median (min–max). Differences between the intervention and control groups were tested using the independent *t*-test or the Mann–Whitney test.
Source: Primary data, 2025.

TABLE 2. Nutrient Intake

Adequacy Level (%)	**Intervention (n=40)**		**Control (n=40)**		**p-value between groups**
	x̄ (±SD)	**Med (Min-Max)**	**x̄ (±SD)**	**Med (Min-Max)**	
Energy Adequacy Level (%)		78.76(58.11-125.49)		75.13(47.45-135.82)	0.273
Protein Adequacy Level (%)	150.38(34.99)			53.00(19.29)	0.001
Fat Adequacy Level (%)		88.81(47.93-114.29)		74.65(43.56-104.76)	0.279
Vitamin A Adequacy Level (%)		0.01 (0.00-41.67)		6.67(0.00-43.91)	0.329
Vitamin B12 Adequacy Level (%)		7.20(0.01-55.60)		4.00(0.00-27.60)	0.859
Iron Adequacy Level (%)	67.13(25.70)		61.63 (20.06)		0.290
Calcium Adequacy Level (%)		88.68(61-256.06)		86.91(57.54-226.67)	0.366
Zinc Adequacy Level (%)		32.46(47.93-114.29)		35.15(7.04-106.42)	0.424

*Values are presented as mean ± SD or median (min–max). Differences between groups were analyzed using the independent *t*-test or the Mann–Whitney test. Nutrient intake was assessed using the 2 × 24-hour dietary recall method and expressed as a percentage of recommended nutrient intake.
Source: Primary data, 2025.

The nutritional adequacy levels of breastfeeding mothers are shown in Table 2. Most nutritional adequacy indicators showed relatively comparable values between the intervention and control groups. There were no significant differences in energy, fat, vitamin A, vitamin B12, iron, calcium, or zinc adequacy. However, protein adequacy levels were significantly higher in the intervention group than in the control group.

Changes in lactoferrin levels in breast milk before and after the intervention are presented in Table 3. At baseline, lactoferrin levels between the intervention and control groups showed relatively similar values. After the intervention period, lactoferrin levels increased in the intervention group, while changes in the control group were relatively small. Within-group analysis showed a significant increase in lactoferrin levels in the intervention group, while no significant changes were found in the control group. Furthermore, the overall

TABLE 3. Changes in Lactoferrin Levels Before and After the Intervention

Variable	Intervention (n=40)		Control (n=40)		p-value between groups
	x̄ (±SD)	Med(Min-Max)	x̄ (±SD)	Med (Min-Max)	
Lactoferrin					
Before	484.90± 18.61		482.68 ±38.38		0.742
After	503.38 ± 24.50		485.18 ±53.49		0.064
Change		24.50(-63.00-73.00)		5.00(-150.00-82.00)	0.032
p value within group		0.001			

*Values are presented as mean ± SD or median (min–max). Within-group comparisons were analyzed using paired *t*-test or Wilcoxon test, while between-group comparisons were analyzed using independent *t*-test or Mann–Whitney test.

Source: Primary data, 2025.

TABLE 4. General Linear Model Analysis of Factors Associated with Lactoferrin Levels

Variable	Coeff-β	95% CI	p-value
Intercept	18.475	(-7.160-29.790)	0.002
(Group 1) Control	-15.975	(-31.977-0.027)	0.050
(Group 2) Intervention			

* regression coefficient; CI = confidence interval. The reference category for the study group is the intervention group. Analysis was performed using the general linear model.

Source: Primary data, 2025.

change in lactoferrin levels was higher in the intervention group than in the control group.

A general linear model analysis was conducted to evaluate factors associated with lactoferrin levels in breast milk. In the initial stage of the analysis, the variable protein adequacy level was entered along with the study group. However, this variable did not show a significant relationship and did not meet the criteria for a confounding variable, so it was not retained in the final model. The final model showed that the study group was associated with lactoferrin levels in breast milk, with respondents in the control group having lower lactoferrin levels compared to those in the intervention group. The results of the final model are presented in Table 4.

This study aimed to evaluate the effect of a local food intervention consisting of tilapia (*Oreochromis niloticus*) and banana blossom (*Musa balbisiana* Colla) on lactoferrin levels in breastfeeding mothers. The results showed that lactoferrin levels increased significantly in the intervention group after the intervention period, while no significant changes were found in the control group. Furthermore, the change in lactoferrin levels between before and after the intervention was also higher in the intervention group than in the control group. General linear model analysis showed that the intervention group was associated with lactoferrin levels after model adjustment. These findings suggest that local food interventions have the potential to increase breast milk bioactive components in breastfeeding mothers.

Lactoferrin is a multifunctional bioactive protein found in breast milk and plays a crucial role in supporting the protection and development of the infant's immune system. This protein has various biological activities, such as antimicrobial, antiviral, anti-inflammatory, and immunomodulatory, which contribute to the body's defense mechanisms during the neonatal period, thus supporting infant health in early life (Donovan, 2016; Hitrova-Nikolova St., Slancheva B., 2021; Telang, 2018). Lactoferrin concentrations in breast milk are known to vary throughout lactation, with the highest levels in colostrum and decreasing in mature milk. However, they can be influenced by various maternal factors such as nutritional status, lactation stage, and the mother's physiological condition (Czosnykowska-Łukacka *et al.*, 2019; Villavicencio *et al.*, 2017). Breastfeeding practices are also influenced by maternal self-efficacy, family support, cultural factors, and health service support, which may indirectly affect maternal nutritional and physiological status during lactation (Yulidasari *et al.*, 2017)

The increased lactoferrin levels found in the intervention group in this study are likely related to the nutritional content of the intervention diet. Tilapia is a source of high-quality animal protein containing various essential amino acids required for protein synthesis. Adequate protein intake in breastfeeding mothers is known to play a role in supporting protein synthesis in mammary tissue, including bioactive proteins secreted through breast milk. Previous research has shown that maternal nutritional composition can influence the composition of bioactive components in breast milk, including lactoferrin (Zen Rahfiludin & Pangestuti, 2020). From a biochemical perspective, the observed increase in lactoferrin levels may be explained by the availability of essential amino acids supplied by Nile tilapia. Lactoferrin is an iron-binding glycoprotein synthesized by epithelial cells and immune cells, and its production depends on adequate amino acid availability for protein biosynthesis (Lea & Azevedo, 2016). Nile tilapia contains high-quality protein rich in essential amino acids such as leucine, lysine, methionine, and valine, which are involved in cellular growth, tissue repair, and protein synthesis (Musa & Aura, 2026). Adequate intake of these amino acids may support the synthesis of immune-related proteins, including lactoferrin, through enhanced metabolic and translational processes (Sah *et al.*, 2021). In addition, tilapia contains micronutrients such as zinc and selenium that contribute to immune regulation, antioxidant defense, and maintenance of cellular homeostasis during the postpartum period (Silveira *et al.*, 2023; Zhang *et al.*, 2026). These nutrients may indirectly facilitate lactoferrin production by supporting optimal immune and metabolic function.

Furthermore, banana blossoms are known to contain various micronutrients and bioactive compounds such as iron, dietary fiber, and phenolic compounds with antioxidant activity. Iron is known to be related to the biological function of lactoferrin as an iron-binding protein that plays a role in defense mechanisms against pathogenic microorganisms (Ruiz-Rico *et al.*, 2025). The content of bioactive compounds in plant foods has also been reported to support immune function and metabolism in breastfeeding mothers, which may indirectly influence the bioactive composition of breast milk. Banana blossom may also contribute through physiological mechanisms associated with lactation and immune regulation. Besides iron, banana blossom contains polyphenols, flavonoids, tannins, and antioxidant compounds that may reduce oxidative stress and inflammatory responses during the postpartum period (Al-Khayri *et al.*, 2022; Chaiwangyen *et al.*, 2022; Mir *et al.*, 2023). Reduced oxidative stress may improve cellular function and create a favorable physiological environment for the synthesis of bioactive proteins (Khelfi, 2024). In addition, banana blossom has traditionally been recognized as a galactagogue food in several Asian countries. Bioactive compounds contained in banana blossom are believed to support prolactin activity and mammary gland function, thereby enhancing lactation performance (Amornlerdpison *et al.*, 2021). The synergistic combination of high-quality protein from tilapia and bioactive phytochemicals from banana blossom may therefore contribute to the increase in lactoferrin levels observed among breastfeeding mothers in the intervention group. This finding is consistent with previous studies in Indonesia reporting that local food-based interventions and traditional herbal preparations may support lactation performance and breastfeeding outcomes among postpartum mothers (Indrayani *et al.*, 2020; Sistiarani *et al.*, 2024).

The results of this study also showed that most indicators of nutritional adequacy of breastfeeding mothers did not differ significantly between the intervention and control groups, except for higher levels of protein adequacy in the intervention group. This finding aligns with the type of intervention provided, namely a diet rich in animal protein. Several previous studies have reported that maternal nutritional intake, particularly protein and certain micronutrients such as iron, zinc, and vitamin B12, may be associated with the concentration of bioactive components in breast milk (Dumrongwongsiri *et al.*, 2021; Gibson *et al.*, 2020). However, the results of the general linear model analysis in this study showed that protein adequacy was not significantly associated with lactoferrin levels after model adjustment. This suggests that increased lactoferrin levels are likely influenced not by a single nutrient but by the interaction of various nutritional and physiological factors in breastfeeding mothers. The General Linear Model analysis demonstrated that the study group remained associated with lactoferrin levels after adjustment. Participants in the control group had lower lactoferrin levels than those in the intervention group, indicating that the local food intervention contributed to the observed increase in lactoferrin levels. These findings suggest that the combination of nutrients and bioactive compounds contained in Nile tilapia and banana blossom may have played an important role in supporting lactoferrin production among breastfeeding mothers.

The findings of this study have practical implications for maternal and child health programs, particularly in regions with abundant local food resources. Strengthening breastfeeding promotion programs should be integrated with nutrition education and culturally appropriate local food interventions, as previous studies have demonstrated the importance of maternal preparedness, community support, and breastfeeding promotion in improving breastfeeding practices (Suryani *et al.*, 2017; Yuliana *et al.*, 2019). The utilization of Nile tilapia and banana blossom represents an affordable, culturally acceptable, and sustainable nutritional strategy to support maternal health during lactation. Considering that both commodities are widely available in Tasikmalaya Regency and many other regions of Indonesia, their incorporation into nutrition education and breastfeeding support programs may contribute to improving maternal nutritional status and enhancing the immunological quality of breast milk. Integrating local food-based interventions with structured breastfeeding support programs may further improve maternal and infant health outcomes, as breastfeeding support modules have been shown to strengthen breastfeeding practices in community settings (Kowara *et al.*, 2024).

In addition to nutritional factors, several other maternal factors have also been reported to influence lactoferrin levels in breast milk, such as parity, breastfeeding frequency, maternal psychological state, and lactation stage (Villavicencio *et al.*, 2017; Wu *et al.*, 2024). Successful breastfeeding is also influenced by social support and community involvement. Previous quasi-experimental studies have shown that peer support interventions can improve breastfeeding duration and prevalence among mothers, highlighting the importance of supportive environments during lactation (Irmawati *et al.*, 2024). In this study, baseline characteristics of respondents, such as maternal age, parity, breastfeeding frequency, breastfeeding duration, and postpartum depression screening scores, showed relatively comparable patterns between the intervention and control groups. This suggests that the differences in lactoferrin levels observed in this study were more likely influenced by the dietary intervention provided than by the respondents' baseline characteristics. This study has several strengths. First, it used an intervention design with a control group, allowing for a more controlled evaluation of changes in lactoferrin levels before and after the intervention. Second, all participants completed the study, ensuring no significant differences.

Conclusion

This study demonstrated that a 14-day intervention using Nile tilapia (*Oreochromis niloticus*) and banana blossom (*Musa balbisiana* Colla) significantly increased lactoferrin levels among breastfeeding mothers. The intervention group showed greater improvements in lactoferrin levels than the control group, and the General Linear Model analysis confirmed the contribution of the intervention to these changes. The findings suggest that local foods rich in high-quality protein, iron, and bioactive compounds may support maternal immunological status during lactation. Given their availability and affordability, Nile tilapia and banana blossom have the potential to be utilized as local food-based nutritional interventions to support maternal and infant health.

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