Investigate the extent impact of body mass index towards origins human primiparous colostrum IgA, IL5 and TNFα profile

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KEYWORDS
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ABSTRACT:
Background
Excess macronutrients provoke the inflammatory mediators release eading a pro-inflammatory microenvirontment and oxidative stress. Consequently, the differential of maternal environments may induce immunologic composition of breast milk may vary considerably. This paper explores the intricate relationship between body mass index (BMI) to human colostrum immunoglobulin A (IgA),tumor necrosis factor 1 alpha (TNF-α), and interleukin 5(IL-5) concentrration variability.
Methods
We collected colostrum samples from 150 primiparous women within 24 h post-partum. Weight and height measurement were conducted to calculate participant BMI. Enzymelinked immunosorbent assay was used to analyse the IgA, TNF-α, IL-5 concentrations.
Result
This work revealed that the highest IgA levels of colostrum milk were confirmed in lactating mother who had BMI >30kg/m2 (IgA 6.05±0.3ng/mL). The highest levels of colostrum TNF-α were confirmed in lactating mothers who had overweight BMI (239.04±0.8ng/mL).Those lactating mothers who had normal range BMI (18.5–22.9 kg/m2) showed the highestIL-5 levels in colostrum (113.81±0.4ng/mL). Moreover, maternal BMI had no significant influence on the IgA, TNF-α and IL-5 levels of colostrum, but Colostrum IgA levels were significantly different based on BMI category (p=0.005) .
Conclusion
The findings suggest that immunoglobulin A composition in breast milk vary depending on maternal BMI category.

1. INTRODUCTION

Colostrum, the first milk produced by mothers after giving birth and cosidered as primary standrat for infant nutrition, is rich in immunoglobulin A (IgA) and plays a crucial role in protecting newborns against infections and supporting their developing immune systems (Garofoli et al., 2023; Akhter et al., 2021; Palmeira & Carneiro-Sampaio, 2016). IgA is the dominant immunoglobulin in colostrum, with concentrations ranging from 2.84 to 8.69 g/L (mean 5.61 g/L) (Sangild et al., 2021; Sánchez-Salguero et al., 2021). IgA in colostrum is essential for infant health, providing protection against infections and supporting immune system development. Interestingly, the levels of IgA in colostrum and breast milk may be associated with various maternal factors, including nutritional status, time postpartum, mode of delivery, parity, and health conditions which significantly affects colostrum composition (Campo et al., 2024; Garofoli et al., 2023; T de Vries et al., 2018).

Malnourished mothers have lower levels of IgG and IgA in their colostrum compared to well-nourished mothers (Miranda et al., 1983). Furthermore, time postpartum is inversely correlated with colostrum total IgA levels, with concentrations dropping rapidly after birth (Munblit et al., 2015). Mode of delivery and maternal parity also influence IgA levels in colostrum (Hirata et al., 2022; Munblit et al., 2015). Interestingly, mothers of infants who later developed atopic symptoms or IgE sensitization had lower concentrations of IgA casein antibodies in their colostrum compared to mothers of non-atopic infants (Savilahti et al., 2005). This suggests a potential link between maternal IgA levels and the development of allergies in offspring. Maternal health conditions, such as gestational diabetes, can affect the immunological composition of colostrum. Mothers with gestational diabetes showed reduced levels of IgA and other immunoreactive proteins in their colostrum (Guerra et al., 2023).

The relationship between obesity, BMI, and inflammatory markers such as IgA, IL-6, and TNF-α in colostrum is complex and not fully consistent across studies. While obesity and increased BMI are generally associated with higher levels of inflammatory markers in serum, the relationship in colostrum appears to be more nuanced, with overweight women showing higher levels of some inflammatory markers compared to both normal weight and obese women.Interestingly, this pattern in colostrum differs from what is typically observed in serum, where obese individuals tend to have higher levels of inflammatory markers. For instance, Khaodhiar et al. (2004) reports that obese subjects had significantly higher serum levels of TNF, IL-6, and other inflammatory markers compared to non-obese subjects. This study aims to investigate the extent of the influence of BMI on changes in IgA, TNFa and IL-5 levels in colostrum of primiparous mothers.

2. METHODS

2.2. Study design

This study used an analytical observational design with a cross-sectional study approach.

2.3. Participant

The research subject recruitment process begins with providing education and explaining the research procedures to earch participant. All first-day primiparous postpartum mothers who gave birth vaginally without complications in the Gianyar Regency area of Bali during the period from October to December 2022 were involved in this research. A non-probability purposive sampling approach was applied to sort participants with the following criteria: Primipara, first day postpartum, vaginal delivery without complications such as obstructed labor, forceps, vacum, and babies born in healthy condition without congenital abnormalities. Mothers with hypertension, diabetes or not in the obesity group as confirmed by BMI measurements, are not HIV sufferers, and have no history of allergies, no history of asthma, rheumatoid arthritis, gastritis and Hodgkin's disease which can affect IgA levels in breast milk. After screening each subject who met the requirements was given a informed consent.

2.4. Socio-demographic and clinical measures

The sociodemographic characteristics studied in this study included age, level of employment, education, weight gain during pregnancy, body mass index,neonatal birth weight and other.

2.5. Sample collection and measurement

Breast milk in first postpartum day was collected in private room, preceded by 15 minutes of breast massage and care by midwife. After centrifuged at 800xg for 10 minutes at 40^0C breast milk samples were placed in 2.5 ml effendof tubes, and stored at -20^0C until assay with ELISA reader (Hawkes et al., 1999; 2002; Bottcher et al., 2003). IL5 examination used human IL5 (ab. 100571) and IgA using human IgA ELISA Kit (no.cat.KA3980 Abnova).

Meanwhile for TNF-α, standards or samples were added to the ELISA plate wells and combined with the pre-coated antibodies. The biotinylated detection antibody is specific for Secretory Immunoglobulin A and Avidin- Horseradish Peroxidase (HRP) conjugate was added to each plate well and incubated. The enzyme-substrate reaction is terminated by the addition of a stop solution. The optical density (OD) is measured spectrophotometrically at a wavelength of 450 nm ± 2 nm..

Data analysis

The results of statistical analysis were presented as mean value and standard deviation. All data analysis was performed out using the STATA 18 software and *p*-values less than 0.05 were considered as statistical significance values in this study.

3. RESULT

A total of 150 participant were enrolled in this investigation, an overall summary of the clinical variables of each study group is presented in Table 1.

It is noticeable from the outcomes and findings from relatedinvestigation that IgA levels in individual colostrum can vary extensively. Mean values of IgA in colostrum recorded range from 1.6 ng/mL reported in this study to 9 g/L reported in a study conducted on healthy Kiwi mother. We analysed the effect of mother's characteristics especiallyat different BMI categories on IgA, TNF-α and IL-5 levels in primiparous colostrum. Maternal BMI had no significant influence on the IgA, TNF-α and IL-5 levels of colostrum, but IL-5 showed statistically significant influence in TNF-α level of colostrum. Result showed that IgA levels of colostrum milk in lactating mother who had BMI >30kg/m^2 were highest (IgA 6.05±0.3ng/mL) compared to lactating women who had other BMI categories (BMI 25-29.9 kg/m^2; IgA: 1.43±0.3ng/mL, BMI 23-24.9 kg/m^2; IgA: 1.56±0.4ng/mL. Normal BMI (18.5-22.9 kg/m^2); IgA:2.58±0.6ng/mL). Colostrum IgA levels were significantly different based on BMI category (*p*=0.005) (Fig.1).

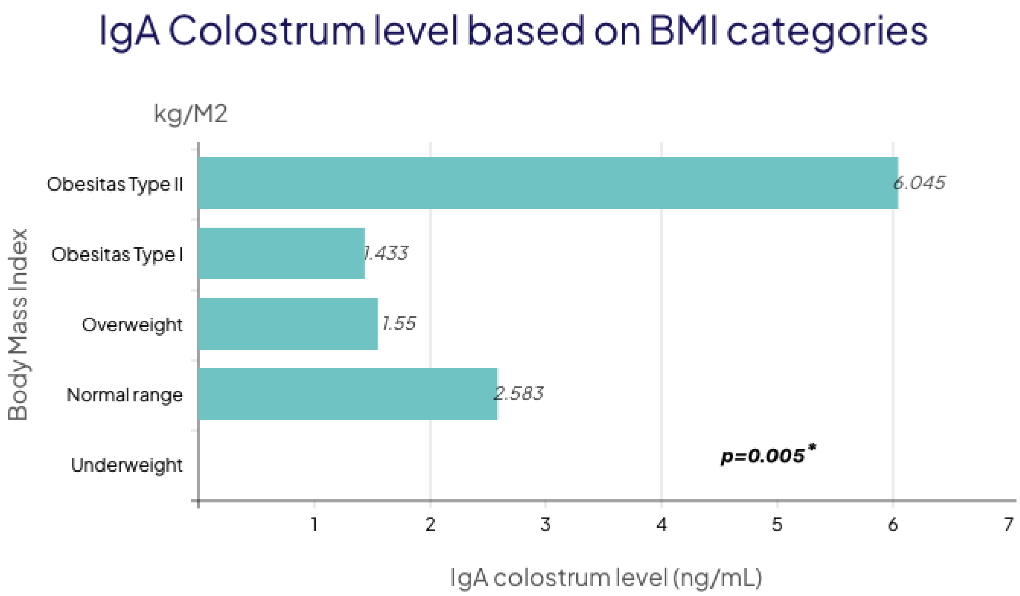

Figure 1. IgA level in primiparous colostrum based on BMI categories

Table 1 Characteristics of study participants between sites of collection.

Indikator	**(n=150)**
Maternal Age (year), mean±SD	25±0.3
Education, n(%)	
Senior high School	59(65.5)
Diploma	15(16.7)
Bachelor	16(17.8)
Job, n(%)	
Housewife	27(30)
Teacher	2(2.2)
Private employees	45(50)
Entrepreneur	16(17.8)
Weight (kg), mean±SD	64,1±0.5
Height (cm), mean±SD	160,2±0,6
Weight gain during pregnancy (kg), mean±SD	13.4±0.3
BMI, n(%)	
Underweight	-
Normal range	12(14.3)
Overweight	31(36.9)
Obesity type I	39(46.3)
Obesity type II	2(2.4)
Birth weight (gr), mean±SD	3264.7±22.5
Immunomodulatory colostrum profile, mean±SD	
IgA	1.6±0.02
IL-5	78.9±0.1
TNF-α	209.9±0.1

Data are expressed as mean ± SD (n=60). Statistical methods used: [1] unpaired t-test. [2] Pearson chi-squared test. *-Results are statistically significant at a level less than 0.05. SD-standard deviation. Abbreviation: kilogram

Moreover, those lactating mothers who had normal range BMI (18.5–22.9 kg/m[2]) showed the highestIL-5 levels in colostrum (113.81±0.4ng/mL) than higher BMI (≥30 kg/m[2]; IL-5: 45.5±0.2ng/mL) or overweight BMI (23–24.9 kg/m[2]; IL-5: 49.41 ± 0.6ng/mL); however, the differences could not reach a significant level (p = 0.456). In this study, it was observed that the highest levels of colostrum TNF-α were confirmed in lactating mothers who had overweight BMI (239.04±0.8ng/mL). Furthermore, the lowest level of TNF-α levels in primiparous colostrum were observed in lactating mothers who had BMI>30kg/m^2 (33.34±0.6ng/mL) compared with normal range BMI (130±0.7ng/mL) or obesity type I BMI (220.23±0.6ng/mL), although the differences of TNF-α levels could not reach a significant level (p=0.584).

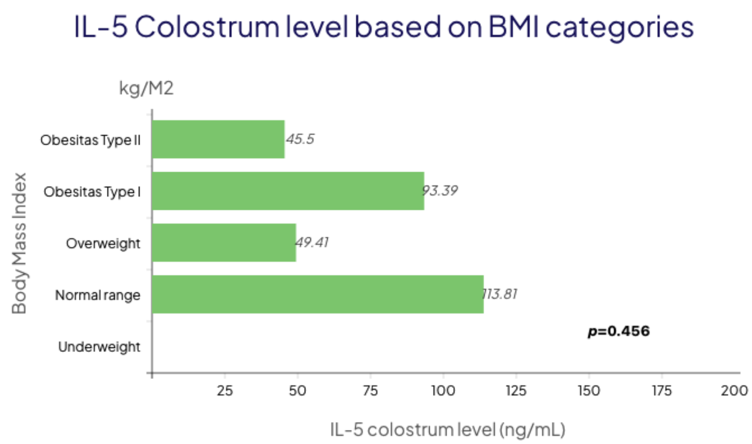

Figure 2. IL-5 level in primiparous colostrum based on BMI categories

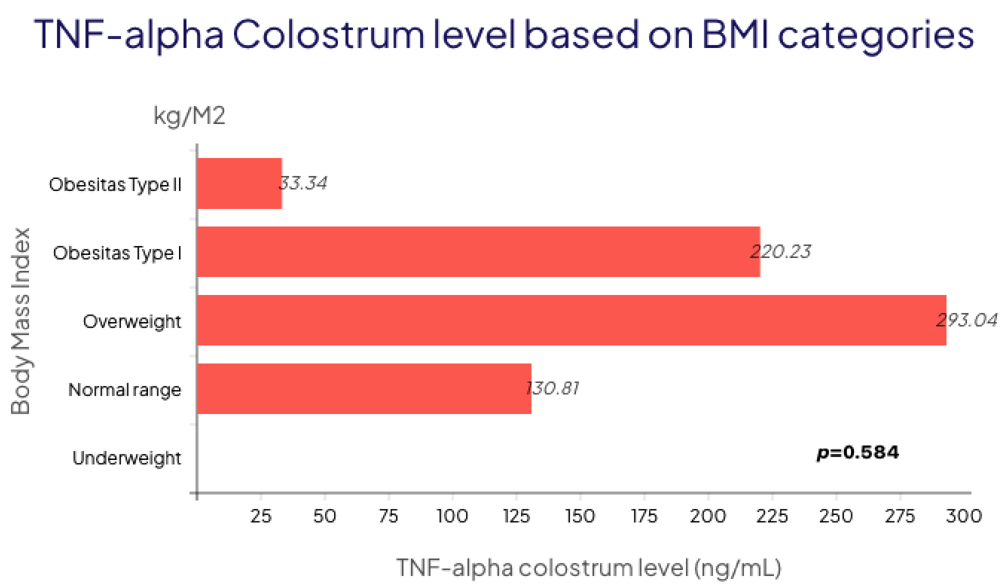

Figure 2. TNF-αlevel in primiparous colostrum based on BMI categories

4. DISCUSSION

The quantitative measurement of the major immunoglobulin, TNF-α and IL-5 in colostrum in first day post-partum periods, it was observed that all three immunomodulatory profile were present in colostrum. The highest concentration of IgA were present only in the lactating mothers who had BMI >30 kg/m^2, while the highest concentration ofIL-5 were present only in the lactating mothers who had normal range BMI. Moreover, the highest concentration of TNF-αwere present only in the lactating mothers who had overweight BMI.Corroborating with this finding, prior study report that higher levels of immunoglobulin A (IgA) and secretory IgA (sIgA) concentrations were found in obese serum and colostrum, respectively (Fujimori et al., 2015).

Previous study reported that breastmilk contains numerous immunologic components including immune cells, antibodies (especially IgA antibodies), pro- and anti-inflammatory cytokines such as TNF, IL-10, and TGF- β (Tomicić et al., 2010). Cytokines are signaling molecules synthesized by most of the nucleated cells and, once transferred via mother colostrum/milk, can cross the intestinal barrier and influence the infant’s immune system. Consequently, the immunologic composition of breast milk may vary considerably maternal related factor including BMI (Fron et al., 2023). Obesity is a condition in which there is an abnormal or excessive accumulation of fat that has the potential to interfere with the continuity of health state (Rubino et al., 2020). Excess macronutrients in adipose tissue provoke the inflammatory mediators release such as TNF-α, and lowering adiponectin synthesis, leading a pro-inflammatory microenvirontment and oxidative stress (Elulu et al., 2017; Hall et al., 2021). Thus, the high IgAs concentrations in maternal colostrum exerts dynamic protection with other bioactive elements, which is particularly needed by preterm infants (Prereira et al., 2023;Sánchez-Salguero et al., 2021).

Overall Inflammation is a complex biological response that significantly influence IgA synthesis. In response to inflammation, immune cells such as B cells are activated. The presence of cytokines and other signaling molecules during inflammation promotes B cell differentiation and proliferation, leading to increased IgA synthesis. IgA is predominantly found in mucosal areas such as the gut, respiratory tract, and salivary glands. During inflammation of these tissues, IgA production can be upregulated to help neutralize pathogens and prevent further infection.Various cytokines associated with inflammation stimulate IgA production. For example, interleukin-5 (IL-5) and transforming growth factor-beta (TGF-β) are particularly important for promoting class switching in B cells to produce IgA (Piñeiro-Salvador et al., 2022).

Furthermore, in prior study of colostrum from women with different body compositions, IL-6 and TNF-α levels were found to be significantly higher in overweight women compared to normal weight and obese women (Murguía-Vázquez et al., 2024). Specifically, IL-6 levels were 55 ± 72.4 pg/mL in overweight women versus 48.1 ± 74.1 pg/mL in normal weight and 28.9 ± 36.2 pg/mL in obese women. TNF-α levels showed a similar pattern, with 58.7 ± 74.9 pg/mL in overweight women compared to 38.6 ± 95.4 pg/mL in normal weight and 52.6 ± 115 pg/mL in obese women (Murguía-Vázquez et al., 2024). Interestingly, this investigating showed that IL-5 concentration were highest in lactating women who had a normal range BMI. Further investigation stated that IL-5 concentrations showed the strongest correlation with adipometrics and may be a possible research and drug target in obesity (Schmidt et al., 2015). Thus, the role of anti-inflammatory cytokines such as IL-5, IL-10 and IL-13 in relation to physical activity in staying metabolically healthy, clarify the reasons for high IL-5 levels in lactating mothers who have a normal BMI range.

5. CONCLUSION

To the best of our knowledge, this is the first study of the main immunomodulator in primiparous colostrum from first day as soos as breastmilk produced startedTherefore, this pioneering study is a stepping stone to further investigate active immunity in human breastmilk. Additional research is necessary to understand the etiology and consequences of the reported alterations in mothers suffering from obesity.

6. AUTHOR CONTRIBUTION

KAK, SW, SA, and NN made substantial contributions to the conception or design of the manuscript. KAK contributed to collecting data, analyzed, interpreted the data, drafting and critical revision of the manuscript. SW, SA, and NN supervised this study. All the authors have read and approved the final version of the manuscript. All authors contributed equally to the manuscript and have read and approved the final version of the manuscript.

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9. DISCLOSURE STEATMENT

The author(s) declare that there are no conflicts of interest.

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