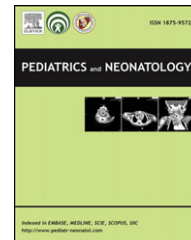




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ORIGINAL ARTICLE

Efficacy of Creamatocrit Technique in Evaluation of Premature Infants Fed With Breast Milk

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Key Words

calorie;
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Background: Most premature babies are discharged with low body weight. Creamatocrit represents the lipid concentration of breast milk. We expected the creatatocrit technique could be applied in the nutrition plan for premature infants who were exclusively fed by human milk. **Methods:** Breast milk samples were obtained from the mothers whose babies were admitted to the neonatal intensive care unit or sick baby room. The breast milk provider was enrolled under the criteria of stable breast milk expression 2 weeks after having given birth. Breast milk was collected for 7 consequent days. Creamatocrit technique and calorie analysis were performed on the processed breast milk samples.

Results: Fourteen pairs of mothers and infants were enrolled in our study. The median gestational age and birth weight were 29 weeks (27–36 weeks) and 1393 g (680–3050 g), respectively. The mean calorie and creatatocrit values for all the 98 breast milk samples were 0.67 kcal/mL and 5.98%, respectively. The linear correlation between creatatocrit value and laboratory-measured calories was found to be calories (kcal/mL) = $0.39 + 0.048 \times \text{crematocrit} (\%)$ ($p < 0.05$).

Conclusion: We established the relation equation of creatatocrit and calories for the first time in Chinese population, which is convenient and accurate for evaluating calories provided for premature infants fed with breast milk.

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1. Introduction

Delayed catch-up in body weight (BW) at discharge is a common problem in premature infants.¹ To provide adequate energy for weight gain under fluid restriction is the critical point in the nutrition management in this group.

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Breast milk, the best food for premature infants, provides not only nutrients but also large amounts of bioactive components that modulate development.^{2,3} Somehow, the lipid content, which acts as the main energy provider of breast milk, is highly variable.⁴ This highlights the necessity of calorie measurement of individualized breast milk for premature infants.

Creatocrit technique, a simple and inexpensive method to estimate the lipid concentration and calories in breast milk, has been extensively used in lactation research.^{2,5,6} The accuracy of creatocrit is important when applied in clinical setting, and there were only data published in Western society and Japan before.^{7,8} On the other hand, breast milk samples were collected and processed in different ways based on different aims in different studies. They might be fresh, processed, foremilk, hindmilk, or composite milk.^{9–11} What actually happens in most neonatal intensive care units is that the baby is fed with human milk expressed by the mother, which has then undergone subsequent processing, being frozen, transported, stored, and reheated before feeding. Also, almost all of the babies are fed with composite milk rather than only foremilk or hindmilk. Furthermore, the hematocrit tube reader, compared with the vernier calipers, is more commonly used in clinical settings.

The aim of this study was to find the equation that can show the relation between the creatocrit value and calories in Chinese population.

2. Methods

Ninety-eight breast milk samples from 14 mothers whose babies were admitted to the neonatal intensive care unit and sick baby room of China Medical University Hospital were obtained during July 2008–December 2008. The subjects were preterm babies who met the following inclusion criteria: (1) gestational age less than 37 completed weeks; (2) free of severe congenital malformations; (3) free of ventilator care [except nasal continuous positive airway pressure (N-CPAP)] at enrollment; (4) free of active infection; and (5) enteral nutrition achieved 100 mL/kg/d. The breast milk providers were enrolled under the criteria of stable breast milk expression 2 weeks after having given birth. Breast milk was collected for the consequent 7 days after informed consent was given. Both foremilk and hindmilk were included. Information on birth weight and gestational age of the infants were available in all. All the cases were enrolled on their 15th day of life.

The samples were frozen in the refrigerator (-40°C) and reheated by flowing warm water ($<60^{\circ}\text{C}$) before the creatocrit technique (modification of Luca's method)¹² and calorie measurement. This means the samples were exactly the same human milk provided to the baby for feeding. Daily BW of the baby was recorded. Each sample was divided into two equal parts after being homogenized by hand shaking for 30 seconds.¹³ One part was for the creatocrit assessment with the creatocrit technique, and the other part was for caloric measurement using a bomb calorimeter (Ika-calorimeter system C4000; Heitersheim, Germany) performed by a technician who was blind to the creatocrit values.

The same investigator (HYL) performed the creatocrit technique for all samples. She withdrew 1 mL breast milk with the capillary tube (75×1.5 mm outside diameter), sealed one end with clay, and then centrifuged the tube for 15 minutes at 12,000 revolutions/min. The tube was placed vertically, and the length of the packed cream layer at the top of the tube was read using the hematocrit tube reader in triplicate immediately after the centrifugation. The average of the three readings was recorded as the value of creatocrit.

This study was approved by the Institutional Review Board of China Medical University Hospital, and informed consent was obtained from all participants in this study.

All correlation analysis were demonstrated by a linear regression line. The spearman's correlation coefficient and significance were tested by Spearman rank correlation analysis (SPSS Inc., Chicago, IL, USA). A p value ≤ 0.05 was considered to be significant.

3. Results

There were 14 infants enrolled in our study. The median gestational age and birth weight were 29 weeks (27–36 weeks) and 1393 g (680–3050 g), respectively. Among these infants, four (28.6%) were very low birth weight and four were extremely low birth weight infants.

The mean number of calories for all the 98 breast milk samples was 0.67 kcal/mL. The linear correlation between creatocrit values and laboratory-measured calories is shown in Figure 1. Linear regression analysis, using creatocrit and caloric value ($n = 98$), produced a relationship of caloric measurement (kcal/mL) = $0.39 + 0.048 \times$ creatocrit (%) ($r_s = 0.81$; $p < 0.05$).

The mean and standard deviation of creatocrit for all the 98 samples were $5.98\% \pm 0.92\%$. The relation between the mean creatocrit value of the same infant during the 7 days and daily BW change during this period of each infant is presented by a scatter plot (Figure 2). The samples with higher mean creatocrit value (over the median value) are presented with solid line, whereas others are presented with dotted line. The higher the mean creatocrit value (solid lines), the higher the BW increment.

The relation between the mean creatocrit value and the BW increment ratio from Day 1 to Day 7, defined as: $(\text{BW D7} - \text{BW D1})/\text{BW D1}$, of each case is shown in Figure 3. Figure 4 shows the linear regression between the mean creatocrit value and BW increment ratio of the selected 12 cases. Case 14 and Case 7 (illustrated as \square and \blacksquare in Figure 3) were excluded because chart review revealed that one had heart failure with large ventricular septal defect, whereas the other had edema possibly because of hypoalbuminemia. The correlation showed a strong and significant relationship between the mean creatocrit value and BW increment in Figure 4 ($r_s = 0.856$, $p < 0.001$).

4. Discussion

Early establishment of adequate enteral nutrition in premature infants not only benefits weight gain but also reduces the use of parenteral nutrition and thus the

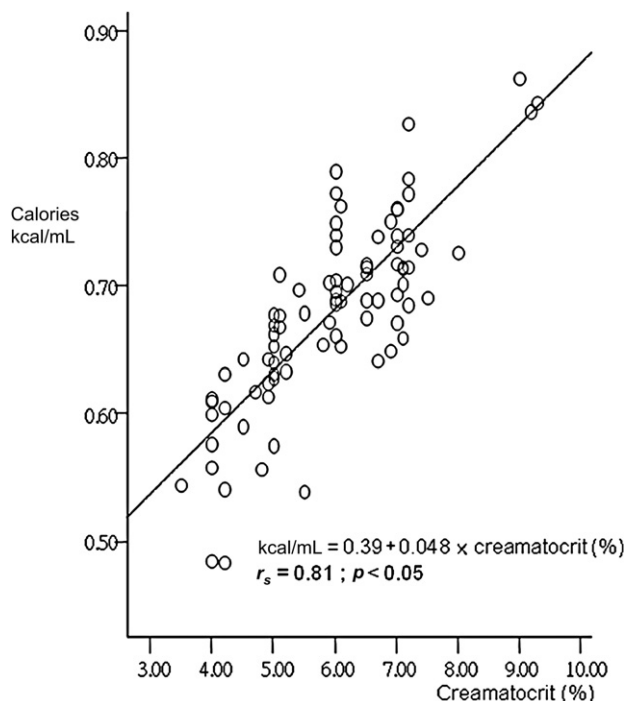


Figure 1 Correlation of calories and creatatocrit. The regression line was calculated by linear regression analysis, and the correlation coefficient and its *p* value were calculated by Spearman correlation analysis.

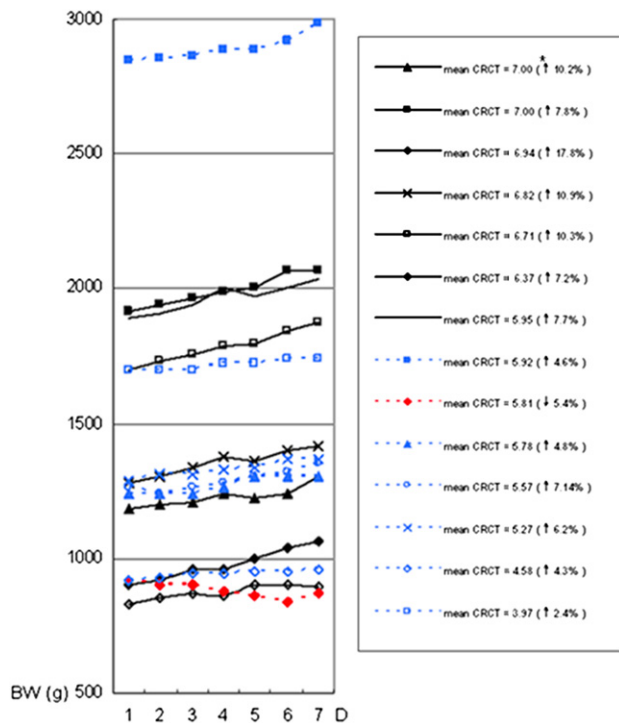


Figure 2 The relation between the mean creatatocrit level of the 7 days and daily weight (g) of each case. The samples with creatatocrit level higher than the median value are presented with solid line, whereas others are presented with dotted line (*body weight increment ratio of each case).

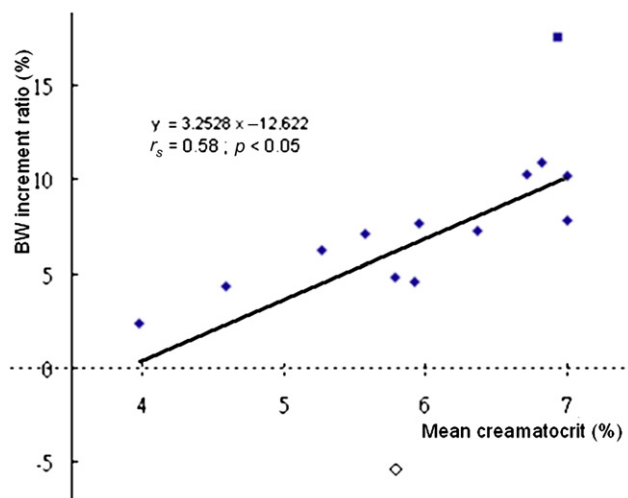


Figure 3 Linear regression between the mean creatatocrit and body weight increment ratio from Day 1 to Day 7 [defined as (BW D7 – BW D1)/BW D1].

associated complications.¹⁴ Creatatocrit, as a tool for measuring lipid component in breast milk, is helpful in estimating if there is adequate energy component to meet the needs of infants in clinical setting.¹⁵

Creatatocrit method has been used as a convenient way in previous studies that related to the breast milk.^{7,8,11,15,16} Yet, no such data were published regarding Chinese population. Our study established the equation that can show the strong relation between the creatatocrit value and corresponding calories for the first time in Chinese population.

Considering the efficacy of using creatatocrit technique in clinical practice, all the samples measured for creatatocrit values and calories were handled in the same process as the breast milk administered to the babies in our study.

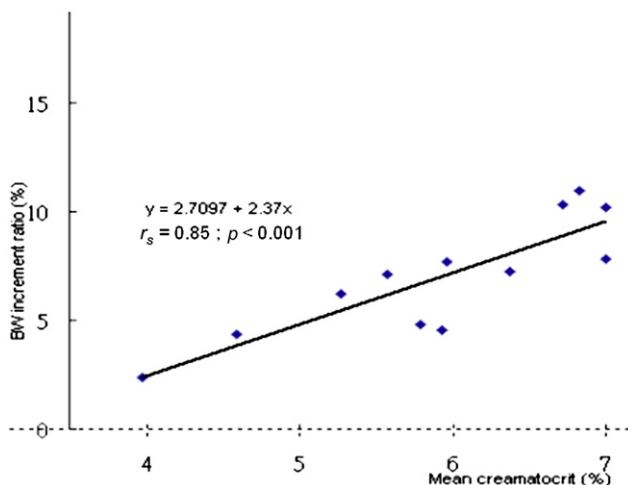


Figure 4 Linear regression between the mean creatatocrit and body weight increment ratio from Day 1 to Day 7 of the 12 selected cases (excluded cases: illustrated as \diamond and \blacksquare in Figure 3).

That means all the samples had been frozen, unfrozen, and warmed before feeding. Also, using breast milk samples exactly the same as the infants were fed for creatocrit analysis and to use the more easily encountered equipment (hematocrit tube reader) for reading of creatocrit value increases the accessibility of nutrition management in daily practice.

Our result shows the mean caloric content for all the 98 breast milk samples was 0.67 kcal/mL, which is similar to the well-known data applied clinically. There was a strong linear relation between the creatocrit value and the calories ($r = 0.81$; $p < 0.05$). We obtained the equation: calories (kcal/mL) = $0.39 + 0.048 \times \text{crematocrit} (\%)$, which is similar to the result from Mizuno et al⁸ [calories (kcal/100 mL) = $38.9 + 5.2 \times \text{crematocrit} (\%)$]. The small difference between the two equations may be explained by the variations in sample management or in reading of the creatocrit by hematocrit reader or vernier calipers.

In our institute, we provide very low birth weight infants intravenous fluid with glucose, electrolytes, and amino acid instead of formulated TPN for parenteral nutrition. Early aggressive enteral nutrition with breast milk feeding is our policy in the nutrition management of premature infants. All the cases were enrolled on their 15th day of life, and all of their nutrition were supported mostly from enteral rather than parenteral when enrolled. As Figure 2 shows, the higher the creatocrit value, the higher the slope of the BW-gain curve.

As shown in Figure 3, there was positive correlation between the creatocrit and changes in the BW increment ratio for the 14 cases. Two infants were noted to be exceptional, and the explanation may lie in the fact that both infants had underlying disease, one had heart failure and one had edema (possibly caused by hypoalbuminemia). After excluding those two infants, the regression analysis obtained a relation as large as 0.856 (Figure 4), which suggests that the relationships between the creatocrit value and the daily BW increment ratio is strong. This implies the possibility of equating creatocrit value with calories of breast milk in daily practice for the caregivers of infants. Although some reports showed that the relation between the creatocrit value and the lipid content is much stronger than that between calories,^{7,9,17} our result revealed such estimated caloric value is useful in clinical practice. Furthermore, this can be applied in the management of nutrition supplement for premature infants,¹⁸ the separation of high-calorie human milk to accelerate weight gain in extremely low birth weight infants,¹⁴ the practice of lactation research related to the mother's nutrition and the content of the human milk, and so forth.

Using 98 human milk samples to analyze the relation between the creatocrit and calorie was reasonable, and the result of our study could serve as a basis for further study in the nutrition management for the preterm babies in Taiwan. Although our study revealed a positive correlation between the creatocrit value and the BW increment ratio, the limitation of this study is that using only 14 infants, it is difficult to declare a good relation between creatocrit value and BW gain. It is warranted to extend the present use of creatocrit technique to examine the relation between the energy content of breast milk and the

infant's weight gain or to examine the relation of the mother's diet and the nutrition component of the breast milk.¹⁹ Also, it could be helpful for clinicians in evaluating breastfed infants.

In conclusion, we have established the relation equation of creatocrit value and calories for the first time in Chinese population, revealing that creatocrit technique is a convenient and accurate method for evaluating the calorie in premature infant fed with breast milk. A new method in this issue has been discussed recently²⁰; however, the creatocrit technique is more convenient and inexpensive for daily practice.

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References

- Gibson A, Carney S, Wales JKH. Growth and the premature baby. *Horm Res* 2006;**65**:75–81.
- Ettyang GA, van Marken Lichtenbelt WD, Oloo A, Saris WH. Serum retinol, iron status and body composition of lactating women in Nandi, Kenya. *Ann Nutr Metab* 2003;**47**:276–83.
- Schack-Nielsen L, Michaelsen KF. Advances in our understanding of the biology of human milk and its effects on the offspring. *J Nutr* 2007;**137**:503S–10S.
- Weber A, Loui A, Jochum F, Bühner C, Obladen M. Breast milk from mothers of very low birth weight infants: variability in fat and protein content. *Acta Paediatr* 2001;**90**:772–5.
- Aksit S, Ozkayin N, Caglayan S. Effect of sucking characteristics on breast milk creatocrit. *Paediatr Perinat Epidemiol* 2002;**16**:355–60.
- Lubetzky R, Littner Y, Mimouni FB, Dollberg S, Mandel D. Circadian variations in fat content of expressed breast milk from mothers of preterm infants. *J Am Coll Nutr* 2006;**25**:151–4.
- Meier PP, Engstrom JL, Murtaugh MA, Vasan U, Meier WA, Schanler RJ. Mothers' milk feedings in the neonatal intensive care unit: accuracy of the creatocrit technique. *J Perinatol* 2002;**22**:646–9.
- Mizuno K, Nishida Y, Sakurai M, et al. Accuracy of creatocrit technique and its efficacy in preterm infants' management. *J Jpn Pediatr Soc* 2006;**110**:1242–6.
- Wang CD, Chu PS, Mellen BG, Shenai JP. Creatocrit and the nutrient composition of human milk. *J Perinatol* 1999;**19**:343–6.
- Vieira AA, Moreira MEL, Rocha AD, Pimenta HP, Lucena SL. Assessment of the energy content of human milk administered to very low birth weight infants. *J. Pediatr (Rio J)* 2004;**80**:490–4 [Article in Portuguese].
- Ogechi AA, William O, Bode-thomas F. Hindmilk and weight gain in preterm very low birth weight infants. *Pediatr Int* 2007;**49**:156–60.
- Lucas A, Gibbs JA, Lyster RL, Baum JD. Creatocrit: simple clinical technique for estimating fat concentration and energy value of human milk. *BMJ* 1978;**1**:1018.
- Jensen RG. Lipids in human milk. *Lipids* 1999;**34**:1243–71.

14. Groh-Wargo S, Sapsford A. Enteral nutrition support of the preterm infant in the neonatal intensive care unit. *Nutr Clin Pract* 2009;**24**:363–76.
15. Mandel D, Lubetzky R, Dollberg S, Barak S, Mimouni FB. Fat and calorie contents of expressed human breast milk in prolonged lactation. *Pediatrics* 2005;**116**:e432–5.
16. Meier PP, Engstrom JL, Zuleger JL, et al. Accuracy of a user-friendly centrifuge for measuring creatinocrits on mothers' milk in the clinical setting. *Breastfeed Med* 2006;**1**:79–87.
17. Lemons JA, Schreiner RL, Gresham EL. Simple method for determining the caloric and fat content of human milk. *Pediatrics* 1980;**66**:626–8.
18. O'Connor DL, Khan S, Weishuhn K, et al. Growth and nutrient intakes of human milk-fed preterm infants provided with extra energy and nutrients after hospital discharge. *Pediatrics* 2008;**121**:766–76.
19. Raiten DJ, Kalhan SC, Hay Jr WW. Maternal nutrition and optimal infant feeding practices: executive summary. *Am J Clin Nutr* 2007;**85**:577S–83S.
20. Corvaglia L, Battistini B, Paoletti V, Aceti A, Capretti MG, Faldella G. Near-infrared reflectance analysis to evaluate the nitrogen and fat content of human milk in neonatal intensive care units. *Arch Dis Child Fetal Neonatal Ed* 2008;**93**: F372–375.