

## Standardization of the methodology for calorimetric analysis of a fermented milk drink

Keicy Sandy Silvestre de Souza<sup>1\*</sup>; Stephanie Pedrosa de Oliveira<sup>1</sup>; Sérgio Murilo Duarte<sup>1</sup>; Igor Viana Brandi<sup>2</sup>; Sérgio Henrique Sousa Santos<sup>2</sup>; Eliane Macedo Sobrinho Santos<sup>3</sup>; Paula Karoline Soares Farias<sup>4</sup>; Hércules Otacílio Santos<sup>3</sup>; Anna Christina de Almeida<sup>2</sup>

### Abstract

The direct calorimetric analysis is the most effective way to obtain caloric value provided by food. Knowing the amount of energy provided by food is essential for the composition of the individuals' diet. Therefore, this paper aimed to standardize a methodology for quantifying the number of calories present in a fermented milk drink with clove essential oil and in a fermented milk drink. Ten mL of each drink were transferred in to triplicate petri dishes and placed in a forced circulation oven 65°C for 24-48 hours to remove moisture to obtain the total solids. These were transferred to a previously tared crucible and inserted into the calorimetric pump until total burning. The results obtained in cal g-1 were converted to Kcal g-1 and finally the amount of Kcal ml -1 of each drink were calculated. The analyzes were submitted to analysis of variance (ANAVA) in a completely randomized design and to the *Tukey* test was used as means of comparison ( $p < 0.05$ ). The Kcal ml -1 values of the analyzed samples did not differ significantly ( $p > 0.05$ ). It can be concluded that the quantification of calories made by this methodology was effective and could later support studies in which it is necessary to calculate with precision the number of calories provided by fermented dairy drinks.

**Keywords:** energy value; calorimetric pump; *Syzygium aromaticum*.

## Padronização da metodologia para análise calorimétrica de uma bebida láctea fermentada

### Resumo

A análise calorimétrica direta é forma mais eficaz de se obter o valor calórico fornecido pelo alimento. Conhecer a quantidade de energia fornecida pelo alimento é imprescindível para composição da dieta dos indivíduos. Dessa forma, esse trabalho visou padronizar uma metodologia para quantificação de calorias presentes em uma bebida láctea fermentada adicionada com óleo essencial de cravo da índia e uma bebida láctea fermentada. Dez mL de cada bebida foram transferidos para placas de petri em triplicada e colocadas em estufa de circulação forçada 65°C por 24-48 horas, para remoção da umidade a fim de obter os sólidos totais. Esses foram transferidos para um cadinho previamente tarado e inserido na bomba calorimétrica até queima total. Os resultados obtidos em cal g-1 foram convertidos em Kcal g-1 e por fim calculado a quantidade de Kcal ml -1 de cada bebida. As análises foram submetidas à análise de variância (ANOVA) em delineamento inteiramente ao acaso e ao teste *Tukey* de comparação de médias ( $p < 0,05$ ). Os valores em Kcal ml -1 das amostras analisadas não se diferiram significamente ( $p > 0,05$ ). Pode-se concluir que a quantificação de calorias feita por essa metodologia foi eficaz e poderá auxiliar posteriormente estudos em que necessite calcular com precisão o número de calorias fornecidas por bebidas lácteas fermentadas.

**Palavras-Chave:** valor energético; bomba calorimétrica; *Syzygium aromaticum*

<sup>1</sup>Mestrando (a) em Produção Animal – ICA/UFMG, Avenida Universitária, 1.000 – Bairro Universitário, cep: 39.404-547, Montes Claros, MG, Brazil.

<sup>2</sup>Docentes do ICA/UFMG, Avenida Universitária, 1.000 – Bairro Universitário, cep: 39.404-547, Montes Claros, MG, Brazil

<sup>3</sup>Docentes do Instituto Federal do Norte de Minas Gerais. Campus Araçuaí. BR 367, km 278, s/n - Zona Rural, Araçuaí - MG, cep: 39600-000, Brasil.

<sup>4</sup>Acadêmica do Programa de Pós-graduação em Ciências da Saúde, PPGCS – Unimontes, Av. Cula Mangabeira, 562 - Santo Expedito, cep: 39401-002, Montes Claros, MG, Brazil

\*Autora para correspondência: [keicysandy@yahoo.com.br](mailto:keicysandy@yahoo.com.br)

## Introduction

Proper nutrition is the basic point to promote and protect human health. The addition of dairy products in the diet, such as fermented milk drink, supplies the energy requirements of at least one of the daily meals of pre-school children, being an alternative food supplement.

The fermented milk drink is produced from a by-product of the dairy industry, whey, having low cost and easy production. Among the chemical preservatives used, the potassium sorbate stands out. However, currently the consumers aim foods that have only natural compounds. Due the clove essential oil has the function of preserving food among its activities, its addition in the fermented milk drink may be beneficial if used in the proper.

In studies that evaluated the nutritional composition of foods, it was observed that the same food presents different amounts of nutrients (Santos *et al.*, 2011). In particular, the fermented milk drinks may vary the energy level to according to the amount of whey and other components used, which can influence the calculation of calories (Grandi; Rossi, 2010).

The concept of calories is always mentioned when discussing nutrition. In a survey of college students, calorie was defined as the energy provided by food that is essential for the body to perform its vital functions. Most respondents said that for healthy eating it is crucial that the food label provide in addition to nutritional information the amount of calories (Fernandes *et al.*, 2015).

Therefore, knowing the amount of energy provided by the food is essential for the composition of the individuals' diet. The energy level of the food should be evaluated by appropriate methods from the energy-providing components such as proteins, fats, carbohydrates, organic acids, and other compounds. The amount of each food component should be converted into energy using indirect calorimetric analyzes, which uses a fixed energy conversion factor for each diet or direct compound, from the burning of food in calorimetric pumps (Food and Agriculture Organization of the United Nations, 2003).

Among the methods used to quantify food calories through energy conversion factors are the Atwater factor, which uses the sum of the protein, lipid and carbohydrate contents in Kcal g<sup>-1</sup> of the food, after being multiplied by four, nine and four respectively (Atwater; Bryant, 1899). However, this method may overestimate the energy value of the food (Novotny; Gebauer; Baer, 2012).

Direct calorimetry is then the most effective way to obtain the caloric value provided by the food. In the scope of nutrition is made by a calorimetric pump that allows to evaluate the energetic content through the heat released by the complete burning of the food (Hackney, 2016).

Therefore, the objective of this study is to standardize a methodology using the IKA C5000® adiabatic calorimetric pump, to quantify the calories in Kcal g<sup>-1</sup> provided by two fermented dairy drinks, one using the chemical preservative, considered conventional and the other added with the essential oil of clove. Because, the adiabatic calorimetric pump, does not quantifies liquids, only solids.

## Standardization of methodology and calorimetric analysis

Four samples of the fermented milk drink with clove essential oil (2 µl ml<sup>-1</sup>) e the fermented milk drink were prepared in the food technology laboratory of the Agrarian Sciences Institute of the Federal University of Minas Gerais, ICA/UFMG.

The caloric value of the fermented milk drinks were determined by direct calorimetry in the Laboratory of Bromatology using the calorimetric pump IKA C5000® according to manufacturer's recommendations ([goo.gl/si9MqD](http://goo.gl/si9MqD)) with adjustments.

With the aid of beaker a ten mL, five mL of the each produced sample was transferred to identified petri dishes in triplicate and placed in the forced circulation oven at 65°C for 24 hours. After 24 hours another five mL were placed in the same petri dishes and stored for 24 hours to remove excess moisture and obtain the total solids.

The total solids were transferred to a crucible, previously tared on an analytical balance, to obtain their weight. In the nickel wire of the calorimetric pump, a cotton thread was pinched and the crucible containing the sample was embedded in the pump so that the cotton yarn was fully in contact with the sample. Once the calorimetric pump was closed and fitted in the calorimeter IKA C5000®.

On the equipment panel, the sample identification data the amount of grams was inserted and the analysis was then started. After complete burning of the total solids of each sample the equipment finished the analysis and provided the calorie value in cal g<sup>-1</sup>.

The total solids value obtained in calories per gram (cal g<sup>-1</sup>) was converted to Kcal g<sup>-1</sup> and finally calculated the amount of Kcal ml<sup>-1</sup> of the fermented milk drink with clove essential oil and of the fermented milk drink. The Kcal values obtained from the three samples were submitted to analysis of variance (ANAVA) by a completely randomized design and, as (p <0.05), the Tukey test of means comparison was performed using the software Rstudio®.

When we analyze the fermented milk drink with clove essential oil (2 µl ml<sup>-1</sup>) e the fermented milk drink

the values in Kcal ml<sup>-1</sup> of the three samples did not present a significant difference ( $p > 0.05$ ) table 1.

Table 1 – Values in Kcal ml<sup>-1</sup> of fermented milk drink with clove essential oil and fermented milk drink obtained from the burning of the total solids in an adiabatic calorimetric pump

Sample identification	Value in Kcal ml <sup>-1</sup>	
	B.O	B.S.O
Sample 1	0.74 Aa	0.72 Aa
Sample 2	0.72 Aa	0.69 Aa
Sample 3	0.73 Aa	0.71 Aa
Sample 4	0.70Aa	0.68 Aa

B.O = fermented milk drink with clove essential oil

B.S.O = fermented milk drink

Means followed by the same capital letters in column and lower case letters on the lines do not differ significantly by the Tukey test at 5% significance level.

Similarly, when compared to the fermented milk drink with clove essential oil with the fermented milk drink with potassium sorbate, within each match, there was no significant difference ( $p > 0.05$ ) as shown in table 1.

In a study conducted by [Grandi and Rossi \(2010\)](#) with fermented milk drinks available in the Brazil, it was observed that the energetic value provided on the label showed differences in more than 80% of the analyzed drinks, which can interfere in the calculation of calories provided by the drink to build the diet of the consumers.

As the studies use the values of protein, fat and carbohydrate according to resolution RDC nº 360/039 ([Brasil, 2003](#)), to calculate the energy value of fermented milk drinks. It can be inferred that the calorimetric analysis performed in our study was more effective, since,

when performing the analysis from the total solids, the calories provided by the drinks did not present significant difference when their samples were evaluated.

## Conclusion

The results of this study demonstrated that the calorimetric analysis performed on this fermented milk drink with clove essential oil and fermented milk drink accurately provided the mean value de Kcal ml<sup>-1</sup>, which may later aid studies in which it is necessary to calculate accurately the number of calories provided by fermented milk drinks.

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