Journal of Veterinary Science and Public Health

J. Vet. Sc. Public Health, v. 7, n. 2, p. 084-096, 2020

ANALYZES OF ALIZAROL, ACIDITY AND FORMALDEHYDE OF UHT MILK COMMERCIALIZED IN THE MUNICIPALITY OF JI-PARANÁ – RONDÔNIA

ANÁLISES DE ALIZAROL, ACIDEZ E FORMALDEÍDO DO LEITE UHT COMERCIALIZADO NO MUNICÍPIO DE JI-PARANÁ - RONDÔNIA

Paulo Henrique Gilio Gasparotto[1,2]; Cíntia Daudt[1]; Flavio Roberto Chaves Silva[1]; Jerônimo Vieira Dantas Filho[1]; Patrícia Geli Feres Marchi[2]; Fernanda dos Anjos Souza[2]; Cinthya de Andrade Gujanswski[2]; Deisiane dos Santos Rodrigues[2]

[1] Universidade Federal do Acre, Programa de Pós-Graduação em Sanidade e Produção Animal Sustentável na Amazônia Ocidental, Rio Branco-Acre, Brasil.
[2] Centro Universitário São Lucas, Curso de Medicina Veterinária, Ji-Paraná-Rondônia, Brasil.
*Autor correspondente: jeronimovdantas@gmail.com
DOI: 10.4025/rcvsp.v7i2.51576

RESUMO: Define-se como leite adulterado aquele que sofreu a subtração de qualquer uma de suas substâncias, bem como a adição de água, de conservantes ou qualquer elemento estranho à sua composição original. Realizou-se análises de nove marcas de leite *Ultra High Temperature* (UHT). De cada marca foram utilizadas seis caixas de leite UHT (1,0 L) de lotes de fabricação distintos, caracterizando, assim seis repetições, totalizando cinquenta e quatro amostras analisadas, sendo essas de lotes de fabricação distintos. Foram realizadas análises de acidez titulável, alizarol, densidade a 15,0ºC e formaldeído. Verificou-se que para a acidez Dornic (ºD), 88,88% das amostras estavam em desconformidade com a legislação. Para o teste do alizarol, 96,29% das amostras analisadas estavam dentro do padrão. Em relação ao teste de densidade a 15ºC, um total de 1,85% das amostras apresentou resultados irregulares. Quanto à presença de formaldeído, 7,40% das amostras foram positivas. Faz-se necessária fiscalização mais eficaz e melhor controle de qualidade na linha de produção e beneficiamento para que o produto seja inócuo ao consumidor.

Palavras-chave: acidez Dornic, conservante no leite, controle de qualidade, Fraude do leite

ABSTRACT: Adulterated milk is defined as that which has been subtracted from any of its substances, as well as the addition of water, preservatives or any element foreign to its original composition. Analyzes of nine Ultra High Temperature (UHT) milk brands were performed. From each brand, six UHT milk boxes (1.0 L) from different batches were used, thus characterizing six repetitions, totaling fifty-four samples analyzed, these being from different batches. Analyzes of titratable acidity, alizarol, density at 15.0ºC and formaldehyde were performed. It was found that for Dornic acidity (ºD), 88.88 % of the samples were not in compliance with the legislation. For the alizarol test, 96.29 % of the samples analyzed were within the standard. In relation to the density test at 15ºC, a total of 1.85 % of the samples presented irregular results. As for the presence of formaldehyde, 7.40 % of the samples were positive. More effective inspection and better quality control in the production and processing line is necessary for the final product to be harmless to the consumer.

Key words: Dornic acidity, milk fraud, preservative of milk; quality control

INTRODUCTION

The milk production chain is one of the most important in the Brazilian agro-industrial complex, presenting one of the largest herds in the world with great potential for supplying milk in the domestic and foreign markets. Milk is among the six most important agricultural products in Brazil, ahead of traditional products such as processed coffee and rice, being essential in providing food and generating jobs and income for the population (CONSOLI and NEVES, 2005).

Milk is the basal source of calcium in human food, whose importance is related to its functions in the human body, as it contributes to the formation of bone tissue, promotes growth, regulates the nervous system and increases resistance to infections (TOMBINI et al., 2012). The physical-chemical characteristics determine the quality of the milk, detecting fraud, such as, for example, adding water or preservatives. Therefore, adulterated milk is defined as that which has been subtracted from any of its substances, except fat for skimmed milk, as well as the addition of water, preservatives or any foreign element to its original composition (MAPA, 2017b).

There are the mandatory official physical-chemical analysis procedures for controlling milk and dairy products, namely: fat, density at 15ºC, total dry extract, defatted dry extract, cryoscopic index, phosphatase, peroxidase and acidity, called quantitative and qualitative (MAPA, 2006). The evaluation of milk quality, through the acidity parameter, through the Dornic degree titration and the alizarol test, which determine the pH, is being widely used, due to the ease and speed of execution and compliance with the current legislation (SILVA et al., 2008; MONTANHINI and HEIN, 2013).

The density test is used as an indicator of fraud and adulteration, and the density of the milk depends absolutely on the dissolved and suspended matter in the researched volume, that is, on the defatted dry extract, for example fat and water. Density is suitable as an indicator for the detection of water residue incorporated during heat treatment or fraud by adding water, since the amount of water present in the milk and the density of the milk are inversely proportional. This is because the density of water is lower when compared to that of milk and the final result tends to approximate the value of water (TAMANINI et al., 2011; MAPA, 2018).

Only the use of sodium citrate, sodium monophosphate, sodium diphosphate and sodium triphosphate as stabilizers in UHT milk is tolerated. However, formaldehyde has been added to milk as a way to preserve the raw material, since this compound has the ability to inhibit the multiplication of microorganisms, masking deficiency problems in the cooling and hygiene system in the milking stage and in equipment involved during processing (MAPA, 1997; SILVA et al., 2016). To be considered fit for human consumption, UHT milk must contain normal sensory characteristics, original fat content for whole milk with 3.0 % fat for standardized milk, stability to the 72.0 % alizarol test, acidity between 14 and 18 ° Dornic and relative density between 1.028 to 1.034, in addition to the absence of adulterants (MAPA, 2018).

The objective of the study was to verify the presence of fraudulent substances in UHT milk marketed in the municipality of Ji-Paraná-RO, through analyzes of alizarol, Dornic acidity, density and presence of formaldehyde.

MATERIAL AND METHODS

The samples of whole UHT milk were purchased from public supermarkets in the municipality of Ji-Paraná-RO. Nine different brands of milk were purchased for this study. Six

boxes of UHT milk from different batches of manufacture were used, characterized as six repetitions, totaling fifty-four samples analyzed.

The analyzes were performed at the Laboratory of Food Analysis and Technology of Products of Animal Origin of the Veterinary Hospital of Centro Universitário São Lucas, Campus of Ji-Paraná-RO. After proper identification of the samples, they were analyzed for the values of titratable acidity, alizarol, density at 15.0ºC and formaldehyde, using as triplicate aliquots.

We used a Dornic acidometer, beaker, 10.0 mL graduated pipettes, Dornic solution (0.111 mol/L sodium hydroxide) and 10 mL of milk with the addition of 4 to 5 drops of 1.0 % (m/v) phenolphthalein neutralized alcohol. The samples were titrated with 0.1 N sodium hydroxide solution until the first persistent strong pink color for approximately 30 seconds, according to the recommendation of Normative Instruction No. 68 of December 12, 2006 (MAPA, 2006). Another way of determining the acidity evaluated was through the alizarol test, which consisted of the rapid and approximate determination of milk acidity by colorimetric method (MAPA, 2006).

This methodology is the combination of the alcoholic test with the colorimetric determination of the pH, through the alizarin indicator, which allows to observe simultaneously the color variation by the pH change and the casein flocculation (TRONCO, 2008). For the test, 10 mL beaker, 10.0 mL graduated pipettes and 75% alcohol-alizarol were used, mixing 2.0 mL of the alcohol-alizarol solution with 2.0 mL of milk and homogenizing. The negative result is to observe, subsequently the homogenization of the sample, brick-red color without the presence of coagulation (MIYOSHI et al., 2016). The yellow color with coagulation indicates milk with an acidity greater than 21°D and the violet color, without coagulation, alkalized or cheated milk with water (MAPA, 2006; COSTA et al., 2008).

For the determination of the presence of formaldehyde, a preservative with antimicrobial action, 10.0 mL beaker, 10.0 mL graduated pipettes, 50.0 % sulfuric acid and 2.0 % iron perchloride were used. The technique consisted of mixing 5.0 mL of milk in 2.0 mL of sulfuric acid and 1.0 mL of iron perchloride, heating the mixture to a boil and observing the presence of a purple or violet color, which represents positivity for formaldehyde, or a yellow color, which indicates negativity for the fraudulent substance (COSTA et al., 2008).

The physical-chemical data obtained in the nine UHT milk brands were evaluated by the Kruskal-Wallis test at the 95 % reliability level, using the Software Action, version 3.3.2 of 2016.

RESULTS AND DISCUSSION

The results obtained through physical-chemical analyzes of titratable acidity, alizarol, density at 15.0 ºC and formaldehyde, described in tables 1 and 2, reveal that all samples (brands) showed non-conformity in at least one of the evaluated items, according to the reference values defined by the Technical Regulation of Identity and Quality of UHT Milk (MAPA, 1997), and the arithmetic mean of the Dornic acidity (ºD) of the evaluated samples was 18.16 ºD (Table 2).

Table 1. Dornic acidity of different brands of UHT whole milk sold in the municipality of Ji-Paraná-RO.

	Samples								
Test	**A1**	**A2**	**A3**	**A4**	**A5**	**A6**	**A7**	**A8**	**A9**
Dornic	17	19	17	19	19	17	17	20	18
Acidity	17	18	18	18	20	16	19	19	18
(ºD)	18	18	18	18	19	18	19	20	19
	17	18	18	18	19	18	17	19	17
	19	19	19	18	19	16	19	18	17
	12	19	20	20	19	17	18	18	20
Total	66.66 C	50 C	66.66 C	66.66 C	100 NC	100 C	50 C	66.66 C	66.66 C
(%)	33.33	50 NC	33.33	33.33			50 NC	33.33	33.33
	NC		NC	NC				NC	NC

C- Conform; NC- Non conform.

With the same objective of this study, Cioglia and Freitas (2017) performed UHT milk analyzes, however, differently from what was observed in this study, 100.0 % of the milk samples were in accordance with the parameters of titratable acidity established by the legislation. The analyzes by Souza et al. (2016), carried out in the state of Mato Grosso, revealed that 148 samples of UHT milk out of 172 submitted to titration in Dornic degrees were above 18 ºD, showing that the high acidity (ºD) has been observed in other regions of the country. When observing the arithmetic mean of each sample evaluated and their respective standard deviations (Table 1), it is noted that 88.88 % of the samples were not in compliance with the legislation.

Table 2. Dornic acidity averages and standard deviations of different brands of UHT whole milk sold in the municipality of Ji-Paraná-RO.

Milk samples	Average and standard deviation	Arithmetic average	Reference value
1	18.66±2.42	18.16*	14 – 18 ºD[1]
2	18.50±0.54		
3	18.33±1.03		
4	18.70±0.44		
5	19.16±0.40		
6	17.00±0.89		
7	18.16±0.98		
8	19.00±0.89		
9	18.16±1.16		

*Arithmetic average; [1]Technical regulation of UHT milk (UAT).

Furthermore, hygienic and refrigeration conditions are fundamental to the quality of milk, as they inhibit the exogenous acidification process resulting from the breakdown of lactose into acids, especially lactic acid, resulting from the fermentation of lactose by microbial metabolism (ROSA et al., 2015). Thus, high acidity is an indication of high bacterial count prior to heat treatment (beneficiation), as there are microorganisms capable of producing thermoresistant enzymes that maintain their activity even after UHT treatment (NOMBERG et al., 2009).

Miyoshi et al. (2016), regarding titratable acidity, observed that 100.0 % of UHT milk samples were in accordance with the minimum parameters. Acidity is an important parameter to assess the conservation status of milk, since milks with acidity outside the physical-chemical standards are considered abnormal and/or unfit for consumption. As the acidity of the milk increases, it indicates shelf life. In addition, the acidity values found may also be due to changes in the composition of the milk, as well as the breed of the animal, the individuality, the colostrum, the lactation period and the sterilization process.

Moretto et al. (2008) evaluated that the titratable acidity of milk can also be influenced according to its content of mineral salts, proteins, the volume of the indicator added and the shade adopted as a turning point in the sample titration by the analyst. Similar results to the previous study are shared by Domareski et al. (2010), that based on the analysis of eight samples of different brands of UHT whole milk, observed that all samples were in accordance with the minimum parameters. Robim et al. (2012), based on the evaluation of 58 samples of various brands of whole UHT milk, found that the averages of the results regarding the physical-chemical parameters of fat, acidity and ESD were in accordance with the legislation.

Table 3 presents the results of the Dornic acidity of the UHT milk brands analyzed. It is observed that there are marks outside the limits established by Decree 9.013 of March 2017 and Normative Instruction 76/77 of 2018. Samples 5, 8 and 2, respectively in decreasing order, presented results superior to the other groups, followed by group 4 which did not differ from groups 3, 7 and 9, while group 6 was the one with the lowest value, not differing from groups 1 and 9 ($P \geq 0.05$).

Table 3. Statistical comparison of the values of Dornic acidity of different brands of UHT whole milk marketed in the municipality of Ji-Paraná-RO.

Milk brands	Dornic
1	18.67±2.42 ^{cd}
2	18.50±0.55 ^{ab}
3	18.33±1.03 ^{abc}
4	18.50±0.84 ^{ab}
5	19.17±0.41 ^{a}
6	17.00±0.89 ^{d}
7	18.17±0.98 ^{bc}
8	19.00±0.89 ^{ab}
9	18.17±1.17 ^{bcd}

Averages followed bye qual letters in the column do not differ significantly ($P>0.05$).

Alizarol is an acidity test, helping to differentiate between salt imbalance and excessive acidity (MAPA, 2006). Of the samples evaluated, 96.29 % were within the standard, being stable and with brick red coloring. While 3.70 % of the samples, although stable, showed purple to violet color in the test, indicating the presence of adulterant in the milk.

According to table 4, there is incompatibility in relation to the studies by Domareski et al. (2010), who observed that only 25.0 % of the analyzed brands were stable to alcohol 72.0 %. On the other hand, the results found by Souza et al. (2016) demonstrated that of the 486 samples analyzed, only 35.4 % were unstable to alcohol 72.0 %.

A result similar to this study was observed by Lima et al. (2012), who, in research on physical-chemical analyzes of UHT whole milk samples marketed in the city of Morrinhos-GO, found alizarol stability in all analyzed samples. In addition, Cioglia and Freitas (2017), who also verified the stability of alizarol in all samples evaluated in research of physical-chemical evaluation of whole UHT milk.

Table 4. Alizarol acidity in different brands of UHT whole milk marketed in the municipality of Ji-Paraná-RO.

	Samples								
Tests	**A1**	**A2**	**A3**	**A4**	**A5**	**A6**	**A7**	**A8**	**A9**
Alizarol	E	E	E	E	E	E	E	EA	E
	E	E	E	E	E	EA	E	E	E
	E	E	E	E	E	E	E	E	E
	E	E	E	E	E	E	E	E	E
	E	E	E	E	E	E	E	E	E
	E	E	E	E	E	E	E	E	E
Total (%)	100 C	100 C	100 C	100 C	100 C	83.33 C 16.66 NC	100 C	83.33 C 16.66 NC	100 C

E- Stable; EA- Stable with adulterant; C- According to; NC- Non conforming.

According to table 5, it can be seen that there was no difference in the means between the different milk brands analyzed in the public supermarkets in the municipality of Ji-Paraná-Rondônia.

Table 5. Alizarol Acidity Averages (Kruskal-Wallis test) in different brands of whole UHT milk marketed in the municipality of Ji-Paraná-RO.

	Alizarol	
Milk brands	**Stable**	**Stable with adulterant**
1	5	0
2	5	0
3	5	0
4	5	0
5	5	0
6	4	1
7	5	0
8	4	1
9	5	0

As for the density test at 15ºC, the arithmetic mean was 1030.96 g/mL (Table 6). However, one sample showed high density and was classified as irregular for this parameter.

Table 6. Averages and standard deviations of density at 15°C (g/mL) in different brands of whole UHT milk marketed in the municipality of Ji-Paraná-RO.

	Average and standard		
Milk samples	**Desviation**	**Arithmetic average**	**Reference value**
1	1032.03±1.06	1030.96*	1028 – 1034[1,2]
2	1029.75±0.71		
3	1030.20±2.06		
4	1032.76±0.44		
5	1032.50±0.00		
6	1029.90±0.32		
7	1030.21±0.53		
8	1031.01±0.78		
9	1030.30±1.74		

*Arithmetic average; [1]Technical regulation on identity and quality of chilled raw milk; [2]Regulation on industrial and Sanitary Inspection of Animal Products.

Regarding the density parameter, there were significant differences ($P<0.05$) between the experimental values found in the different samples in this study (Table 7). These results corroborate those obtained by Rosa et al. (2015), where the values for density in the

comparisons between samples showed significant differences (P<0.05). However, the differences found in this parameter do not characterize the existence of abnormalities in the values found, since 100 % of the samples complied with the legislation.

As in the studies by Robim (2011) and Souza et al. (2016), no samples showed a density lower than 1028. Also, Tamanini et al. (2011), when evaluating 33 samples of UHT milk, observed that only one of the samples was out of the standard for density.

Table 7. Density results (g/mL) in different brands of UHT whole milk marketed in the municipality of Ji-Paraná-RO.

	Samples								
Testes	**A1**	**A2**	**A3**	**A4**	**A5**	**A6**	**A7**	**A8**	**A9**
Density	1031.5	1031.0	1031.9	1033.0	1032.5	1029.6	1030.0	1030.2	1030.6
at 15ºC	1031.5	1030.0	1031.9	1033.0	1032.5	1029.6	1031.0	1031.1	1029.6
(ºQ)	1032.5	1030.0	1028.0	1033.0	1032.5	1029.6	1030.0	1031.1	1030.4
	1031.5	1029.0	1027.2	1031.9	1032.5	1030.2	1030.0	1031.1	1030.9
	1031.5	1029.0	1031.1	1032.7	1032.5	1030.2	1030.0	1031.4	1029.9
	1034.0	1029.0	1031.1	1033.0	1032.5	1030.2	1030.0	1032.2	1030.4
Total	100 C	100 C	83.33 C	100 C	100 C	100 C	100 C	100 C	100 C
(%)			16.66 NC						

E- Stable; EA- Stable with adulterant; C- According to; NC- Non conforms.

There are several causes of density variations that do not affect the quality of the milk and are considered normal, such as the composition of the milk in relation to the fat content, the protein value and its temperature at the time of determination.

Table 8 summarizes the results regarding the milk density of the UHT milk brands analyzed. It is observed that all brands are within the limits established by Decree 9013 of March 2017 and Normative Instruction 62 of 2011. Brands 4, 5 and 1 (Table 8), respectively, in decreasing order, presented superior results to the other groups, followed by group 8, which did not differ from groups 9, 7 and 3, while group 2 was the one with the lowest level, not differing from groups 6 and 7 (P≥0.05).

Table 8. Density results (g/mL) (Kruskal-Wallis test) in different brands of UHT whole milk marketed in the municipality of Ji-Paraná-RO.

Milk brands	Density at 15ºC (ºQ)
1	1032.03±1.06 [a]
2	1029.75±0.71 [d]
3	1030.20±2.06 [bc]
4	1032.77±0.44 [a]
5	1032.50±0.00 [a]
6	1029.90±0.33 [cd]
7	1030.22±0.53 [bcd]
8	1031.02±0.78 [b]
9	1030.97±1.75 [bc]

Averages with the same letters do not differ significantly (P>0.05).

Among the abnormal causes of density variation, we can highlight the frauds for reconstitution and presence of serum or water (MENDES et al., 2010). Regarding density at 15.0ºC, Miyoshi et al. (2016) observed a variation from 1029 to 1031 g/mL, and IN nº 76/2018 establishes the density between 1028 to 1034 g/mL for chilled raw milk. In determining density, temperature must be taken into account, as it has a notable influence on body volume, adding or decreasing the denominator (GASPAROTTO, 2017).

Robim et al. (2012), analyzing the density at 15.0ºC of 58 samples of different brands of whole UHT milk, observed results similar to those of this study, with an average of 1029 to 1030 g/mL. Likewise, Domareski et al. (2010), checking the density at 15.0ºC of eight samples of several brands of whole UHT milk, observed variations from 1028 to 1029 g/mL. Cioglia and Freitas (2017) found that 2.7 % of the samples showed density at 15.0ºC below 1028 g/mL, a value close to that of the present study. Regarding the evaluation of the presence of formaldehyde, 4 out of 54 samples, corresponding to 7.40 %, were positive for the presence of the preservative.

Fernandes and Maricato (2010) analyzed 50 samples of raw milk from a dairy in Bicas-MG and observed that in 100.0 % of them there was no detection of formaldehyde. Likewise, Rosa et al. (2015) did not identify the presence of the preservative in the twelve samples of UHT milk from different brands sold in Erechim-RS. The same authors explained that the addition of preservatives, although prohibited, is carried out with the purpose of masking the hygienic-sanitary quality of milk, eliminating microorganisms and increasing the useful life of the product by the addition of urea, that has formaldehyde in its composition. The authors also described that the addition of alkaline solutes to prolong the conservation or decrease the acidity of milk, such as bicarbonates, formaldehyde, boric acid, hydrogen peroxide, potassium bichromate, hypochlorites and salicylic acid, in spite of fraud, have been used as a method of conservation.

Souza et al. (2010), researching the presence of preservatives in 100 samples of UHT milks produced in six Brazilian states (Paraná, Rio Grande do Sul, São Paulo, Minas Gerais, Rio de Janeiro and Goiás) detected the presence of formaldehyde in 44 (44.0 %) of the samples, followed by hydrogen peroxide in 30 (30.0 %) and chlorine in 12 (12.0 %) of the analyzed samples. However, Correa and Campos (2015) analyzed 11 samples (Brands) of UHT milk, and found that 100.0 % of the samples were in compliance with the legislation, different from what was observed in this work.

Formaldehyde and hydrogen peroxide are used in milk fraud aimed at paralyzing microbial activity. Milks with high microbial load show altered pH and, consequently, high Dornic acidity, diagnosed through platform tests performed during reception, which causes refusal of milk by the dairy (ROSA-CAMPOS, et al., 2011). Firmino et al (2010), in a study with raw milk from expansion tanks in the region of Rio Pomba, Minas Gerais, found the presence of formaldehyde and nitrate in 13.0 % and 40.0% of the samples, respectively.

The presence of formaldehyde in whole UHT milk shows that there was a fraud in this food, in order to preserve it, avoiding the action of microorganisms, being a serious aggravating factor for public health, since formaldehyde has a great carcinogenic potential in humans, can cause damage to dairy products due to the decrease in both yield and nutritional value, can change the product quality and, mainly, is a risk to consumers due to the addition of substances harmful to health. Some operations carried out by the Ministry of Agriculture, Livestock and Supply are worth mentioning in Brazil, as was the case in 2013 of the Operation “Leite Compensado” that took place in Rio Grande do Sul. In order to increase the volume of milk, untreated water was added, and to reconstitute the milk patterns of protein, agricultural urea containing formaldehyde was added, the latter being the primary substance approved by the World Health Organization. Approximately 100 million liters of milk were contaminated with formaldehyde in one year, and the profit from fraud was R$ 9.5 million (GRIZOTTI, 2013).

The National Cancer Institute (INCA) characterizes formaldehyde as a gas produced worldwide, on a large scale, from methanol. It is also known as formalin, methyl aldehyde, methylene glycol, methylene oxide, methanal, formaldehyde 40, morbicide, formalite (INCA, 2016). This substance can compromise the organoleptic characteristics and the nutritional value of milk (EVANGELISTA, 2000), as well as bringing risks to the health of the consumer of this product. As of July 2004, the International Agency for Research on Cancer - IARC classified

this compound as carcinogenic (Group 1), tumorogenic and teratogenic for producing effects on human reproduction (Technical Report ANVISA 53/2013).

CONCLUSIONS

The physical-chemical analyzes used under the different brands of UHT whole milk sold in public supermarkets in the municipality of Ji-Paraná-RO, were shown on average within the standards required by law. The main problems found in UHT milk were high acidity and the presence of formaldehyde. These non-conformities may be linked to the conservation of a product with a high microbial load and the addition of preservative to avoid the acidity of the product. Thus, there is a need to improve guidance and inspection of milk and dairy producers, to detect flaws and obtain a quality product suitable for human consumption.

ACKNOWLEDGMENTS

Acknowledgments to the Laboratory of Food Analysis and Technology of Animal Products of the Veterinary Hospital of Centro Universitário São Lucas.

CONFLICTS OF INTEREST

The authors declare that there is no conflict of interest.

REFERENCES

BELOTI, V. et al. Determinação do parâmetro de crioscopia para leite UHT. **Semina: Ciências Agrárias**. v. 36, n. 5, p.3181-3188, 2015. Available from: < http://www.uel.br/revistas/uel/index.php/semagrarias/article/view/19134>. Accessed in: 10 jan. 2019.

BIASI, T.I. **Controle de Qualidade de Leite e Derivados**. 2.ed. São Paulo-SP: Cap-lab, 2012. 368p.

BRASIL. Ministério da Agricultura, Pecuária e Abastecimento. Decreto nº 9.013 de 29 de março de 2017. **Regulamenta a Lei nº 1.283, de 18 de dezembro de 1950, e a Lei nº 7.889, de 23 de novembro de 1989, que dispõem sobre o Regulamento da Inspeção Industrial e Sanitária de Produtos de Origem Animal**. Diário Oficial da União, Brasília, DF, p. 3, 30/03/2017. Seção 1.

BRASIL. Ministério da Agricultura, Pecuária e Abastecimento. Decreto nº 9.013 de 29 de março de 2017. **Regulamenta a Lei nº 1.283, de 18 de dezembro de 1950, e a Lei nº 7.889, de 23 de novembro de 1989, que dispõem sobre o Regulamento da Inspeção Industrial e Sanitária de Produtos de Origem Animal**. Diário Oficial da União, Brasília, DF, p. 3, 30/03/2017. Seção 1.Capítulo III-Art. 256.

BRASIL. Ministério da Agricultura, Pecuária e Abastecimento. Secretaria de Defesa Agropecuária. Instrução Normativa nº 68 de 12 de dezembro de 2006. **Métodos Analíticos Oficiais Físico-Químicos para Controle de Leite e Produtos Lácteos, em conformidade com o anexo desta Instrução Normativa, determinando que sejam utilizados nos Laboratórios Nacionais Agropecuários**. Diário Oficial da União, Brasília, DF, p.8, 14/12/2006. Seção 1.

BRASIL. Ministério da Agricultura, Pecuária e Abastecimento. Instrução Normativa n° 76 de 26 de novembro de 2018. **Aprova o Regulamento Técnico de Produção, Identidade e Qualidade do Leite tipo A, o Regulamento Técnico de Identidade e Qualidade de Leite Cru Refrigerado, o Regulamento Técnico de Identidade e Qualidade de Leite Pasteurizado e o Regulamento Técnico da Coleta de Leite Cru Refrigerado e seu Transporte a Granel**. Diário Oficial da

União, Brasília, DF, p.9, 30/11/2018. Seção1.
BRASIL. Ministério da Agricultura, Pecuária e Abastecimento. Instrução Normativa n° 77 de 26 de novembro de 2018. **Critérios e Procedimento para a Produção, Acondicionamento, Conservação, Transporte, Seleção e Recepção do Leite Cru em Estabelecimentos Registrados no Serviço de Inspeção Oficial.** Diário Oficial da União, Brasília, DF, p.10, 30/11/2018. Seção 1.
BRASIL. Ministério da Agricultura, Pecuária e Abastecimento. Portaria nº 370 de 04 de setembro de 1997. **Norma para a Inclusão do Citrato de Sódio no Regulamento Técnico para Fixação de Identidade e Qualidade do Leite UHT**. Diário Oficial da União, Brasília, DF, p.19700, 08/09/1997. Seção 1.
BRASIL. Portaria INMETRO nº 220, de 23 de julho de 2009. **Autoridade Brasileira de Monitoramento da Conformidade aos Princípios das Boas Práticas de Laboratório**. Brasília: INMETRO, 2018.
CASTANHEIRA, G. C. A. **Controle de Qualidade de Leite e Derivados**. 2.ed. São Paulo-SP: Cap-lab, 2012. 368 p.
CIOGLIA, C. R.; FREITAS, M. T. Qualidade microbiológica de leites UHT comercializados na cidade de Ouro Preto, MG. **Brazilian Journal of Food Research**, v.8, n.4, p.74-88, 2017. Available from: <https://www.repositorio.ufop.br/bitstream/123456789/10877/1/ARTIGO_QualidadeMicrobiol%C3%B3gicaLeites.pdf>. Accessed in: 30 jun. 2019.
CONSOLI, M. A.; NEVES, M. F. **Estratégias para o leite no Brasil**. São Paulo: Atlas, 2006.
CORDS, B. R. et al. Cleaning and Sanitizing in Milk Production and Processing. In: MARTH, E. H.; STEELE, J. L. **Applied dairy microbiology**. 2.ed. New York: Marcel Dekker, 2001, p.547-587.
CORREA, T. F.; CAMPOS, S. A. S. Presença de antibióticos, conservantes e reconstituintes em leite UHT e pasteurizado. **Revista Demetra**, p.289-298, 2015. Available from: <https://www.e-publicacoes.uerj.br/index.php/demetra/article/view/14843>. Accessed in: 13 mar. 2019.
COSTA, F. A.; CAVALCANTI, J. E. A.; MENDES, F. F.; BUENO, N. P.; TOYOSHIMA, S. H. **Diagnóstico Socioeconômico da Região do Circuito do Queijo**. Belo Horizonte: SEBRAE/FUNARBE, 2008.
DOMARESKI, J. L. et al. Avaliação físico-química e microbiológica do leite UHT comercializado em três países do Mercosul (Brasil, Argentina e Paraguai). **Archivos Latinoamericanos de Nutrición**, v.60, n.3, p.261-269, 2010. Available from: < https://www.alanrevista.org/ediciones/2010/3/art-8/>. Accessed in: 10 jan. 2019.
EVANGELISTA, J. **Tecnologia de Alimentos**. 2 ed. São Paulo: Atheneu, 2000. p.577-578.
FERNANDES, V. G.; MARICATO, E. Análises físico-químicas de amostras de leite cru de laticínios em Bicas-MG. **Revista do Instituto de Laticínios Cândido Tostes**, v.65, n. 375, p. 3-10, 2010. Available from: <https://www.revistadoilct.com.br/rilct/article/view/129>. Accessed in: 30 mai. 2019.
FIRMINO, F. C. et al. Detecção de fraudes em leite cru dos tanques de expansão da região de Rio Pomba, Minas Gerais. **Revista do Instituto de Laticínios Cândido Tostes**, v.65, n.376, p.5-11, 2010. Available from: < https://www.revistadoilct.com.br/rilct/article/view/136>. Accessed in: 19 set. 2019.
GASPAROTTO, P. H. G. **Avaliação do leite UHT comercializado no município de Ji-Paraná-Rondônia**. 2017. 42p. Dissertação (Mestrado em Produção Animal) – Universidade Brasil, São Paulo, 2017.
GARCIA, R. V. et al. Estudo de rótulos de leite fermentado comercializados no município de João Pessoa – PB. **Revista Verde de Agroecologia e Desenvolvimento Sustentável**, v.7, n. 1, p. 15-18, 2012. Available from: <https://www.gvaa.com.br/revista/index.ph

p/RVADS/article/view/744>. Accessed in: 14 jul. 2019.

GRIZOTTI, G. **Ministério Público faz operação contra adulteração de leite no RS**. 2013. Available from: <http://g1.globo.com/rs/rio-grande-do-sul/noticia/2013/05/ministerio-publico-faz-operacao-contra-adulteracao-de-leite-no-rs.html> . Accessed in: 10 fev. 2020.

INCOTERM. **Instruções de uso para lacto-Densímetro**. INCOTERM-Indústrias de Termômetros. Porto Alegre-RS. 2008. Available from: <https://www.irmaoshaluli.com.br/media/wysiwyg/PDF/5783_MANUAL_LACTO-DES_METRO.pdf>. Accessed in: 13 jul. 2019.

INCA. Instituto Nacional do Câncer - Ministério da Saúde. In: **Formol ou Formaldeído**. Available from: <http://www1.inca.gov.br/conteudo_view.asp?id=795>. Accessed in: 10 feb. 2020.

MENDES, C. G. et al. Análises Físico-químicas e pesquisa de fraude no leite informal comercializado no município de Mossoró, RN. **Ciência Animal Brasileira**, v.11, n.2, p.349-356. 2010. Available from: <https://www.revistas.ufg.br/vet/article/view/1146>. Accessed in: 11 jul. 2019.

MILANI, M. P. **Qualidade do leite em diferentes sistemas de produção, anos e estações climáticas no Noroeste do Rio Grande do Sul**. 2011. 67 f. Dissertação (Mestrado em Ciência e Tecnologia de Alimentos) - Universidade Federal de Santa Maria. Santa Maria-RS, 2011.

MIYOSHI, L. Y. et al. Rotulagem de leites UHT comercializados no varejo. **Revista do Instituto de Laticínios Cândido Torres**, v.71, n.1, p.19-25, 2016. Available from: < https://www.researchgate.net/publication/315636303_ROTULAGEM_DE_LEITES_UHT_COMERCIALIZADOS_NO_VAREJO>. Accessed in: 21 jun. 2019.

MONTANHINI, M. T. M.; HEIN, K.K. Qualidade do leite cru comercializado informalmente no município de Piraí do Sul, estado do Paraná, Brasil. **Revista do Instituto Laticínios Cândido Tostes**, v.68, n.393, p.10-14, 2013. Available from: <https://www.gvaa.com.br/revista/index.php/REBAGRO/article/view/4708>. Accessed in: 13 jul. 2019.

MORETTO, et al. **Introdução à ciência de alimentos**. 2.ed. ampliada e revisada. Florianópolis: UFSC, 2008. 255p.

NOMBERG, M. F. B. L. et al. Bactérias psicotróficas e atividade proteolítica no leite cru refrigerado. **Acta Scientiae Veterinariae,** v.37, n.2, p.157-163, 2009. Available from: <http://www.ufrgs.br/actavet/37-2/art825.pdf>. Accessed in: 21 jul. 2019.

PEDROSA, D. **Controle de Qualidade de Leite e Derivados**. 2º Edição. São Paulo-SP: Cap-lab; 2012. 368 p.

PZL EQUIPAMENTOS. **Manual de operação de Crioscópio PZL 900**. Revisão 1.0.Londrina-PR.2011.PZL Engenharia.

REZER, A. P. S. **Avaliação da Qualidade Microbiológica e Físico-Química do leite UHT integral comercializado no Rio Grande do Sul**. 2010. 73p. Dissertação (Mestrado em Ciência e Tecnologia de Alimentos) – Universidade Federal de Santa Maria, Santa Maria-RS, 2010.

ROBIM, M. S. et al. Pesquisa de fraude no leite UAT integral comercializado no estado do Rio de Janeiro e comparação entre os métodos de análises físico-químicas oficiais e o método de ultrassom. **Revista do Instituto de Laticínios Cândido Tostes**, v.67, n.389, p.43-50, 2012. Available from:<http://www.ufrgs.br/actavet/37-2/art825.pdf>. Accessed in: 30 jan. 2019.

ROBIM, M. S. **Avaliação de diferentes marcas de leite UAT comercializadas no Estado do Rio de Janeiro e o efeito da fraude por aguagem na fabricação, composição e análise sensorial de iogurte**. 2011. 98p. Dissertação (Mestrado em Higiene Veterinária) - Universidade Federal Fluminense, Niterói-RJ, 2011.

ROSA-CAMPOS, A. A. et al. Avaliação físico-química e pesquisa de fraude em leite pasteurizado integral tipo C produzido na região de Brasília, Distrito Federal. **Revista do Instituto de Laticínios Cândido**

Tostes, v.66, n.379, p. 30-4, 2011. Available from: <https://www.revistadoilct.com.br/rilct/article/view/159>. Accessed in: 11 jan. 2020.

ROSA, L. S. et al. Avaliação da qualidade físico-química do leite ultrapasteurizado comercializado no município de Erechim-RS. **Revista Vigilância Sanitária em Debate**: Sociedade, Ciência & Tecnologia, v.3, n.2, p.99-107, 2015. Available from: < https://pt.scribd.com/doc/286177787/Avaliacao-da-qualidade-fisico-quimica-do-leite-ultra-pasteurizado-comercializado-no-municipio-de-Erechim-RS >. Accessed in: 30 jan. 2019.

SILVA, D. G. et al. Qualidade do leite UHT comercializado na cidade de Itaqui-RS. In: Salão internacional de ensino, pesquisa e extensão, 2016. **Anais do Salão Internacional de Ensino, Pesquisa e Extensão**, v.8, n.2, 2016. Available from: < http://seer.unipampa.edu.br/index.php/siepe/article/view/18269>. Accessed in: 30 jan. 2019.

SILVA, M. C. D. et al. Caracterização microbiológica e físico-química de leite pasteurizado destinado ao programa do leite no Estado de Alagoas. **Revista Ciência e Tecnologia de Alimentos**, v.28, n.1, p.226-230, 2008. Available from: <http://www.scielo.br/pdf/cta/v28n1/31.pdf>. Accessed in: 21 ago. 2019.

SOUZA, H. P. M. et al. Ocorrência do leite instável não ácido (LINA) na região norte do Mato Grosso. **Revista Ciência e Tecnologia de Alimentos,** v.71, n.1, p.38-42, 2016. Available from: <https://www.revistadoilct.com.br/rilct/article/download/513/400>. Accessed in: 22 jun. 2019.

SOUZA, A. H. P. et al. Avaliação físico-química do leite UHT e pasteurizado comercializado na cidade de Londrina-PR. **Revista Brasileira de Pesquisa em Alimentos**, v.1, n.1, p.39-42, 2010. Available from:<https://visaemdebate.incqs.fiocruz.br/index.php/visaemdebate/article/download/438/223>. Accessed in: 21 jan. 2019.

TAMANINI, R. et al. Contribuição ao estudo da qualidade microbiológica e físico-química do leite UHT. **Revista do Instituto de Laticínios Cândido Tostes**, v.66, n.382, p.27-33, 2011. Available from: <https://www.revistadoilct.com.br/rilct/article/view/179>. Acessed in: 24 mar. 2019.

TOALDO, I. M. et al. Multiclass analysis of antibacterial residues in milk using RP-liquid chromatography with photodiode array and fluorescence detection and tandem mass spectrometer confirmation. **Talanta**, v.99, p.616-624, 2012. Available from: < https://www.sciencedirect.com/science/article/pii/S0039914012005140>. Accessed in: 22 abr. 2019.

TOMBINI, H. et l Consumo de leite de vaca entre agricultores. **Revista Alimentos e Nutrição**, v.23, n.2, p. 267-274, 2012. Available from: <http://serv-bib.fcfar.unesp.br/seer/index.php/alimentos/article/viewArticle/2013>. Accessed in: 23 mai. 2019.

TRONCO, V. M. **Manual para inspeção da qualidade do leite**. Santa Maria: Editora UFSM, 3º ed., p. 203. 2008.