REVIEW ARTICLE Bhartiya Krishi Anusandhan Patrika, Volume 37 Issue 1: 71-75 (March 2022)

Innovation Techniques to Assess Adulterated Ghee: A Review

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10.18805/BKAP452

ABSTRACT

Adulteration in dairy products is not only serious trouble to mortal health but also causes profitable losses. Ghee, the most precious fat, is vented at a high price over other fats and oil. Unethical vendors take advantage of this by mixing ghee with inexpensive fats or oils. Adulteration of ghee could be estimated through its physical-chemical constants such as Butyro-Refractometer reading, Reichert-Meissl value, Polenske value, Iodine value, saponification value, which are time-consuming, Therefore, researchers nowadays adopted novel techniques. The addition of coconut oil in ghee is detected using Attenuated total reflectance-Fourier transform infrared spectroscopy (ATR-FTIR) and chemometrics. Analysis of the adulteration in cow ghee with soybean oil by olfactory machine system, electronic nose (E-nose) tool is used as a rapid technique. Analysis of adulteration in cow ghee with vanaspati by using an image analysis technique called particle analysis and colour measurement and a rapid and accessible protocol based on β-sitosterol using Reversed-phase thin layer chromatography (RP-TLC) is developed to check the purity of milk fat. DPPH (2, 2-diphenyl-1-picrylhydrazyl) based chromogenic assay is a rapid method that has been used to identify the presence of palm oil in ghee.

Key words: Adulterants detection, Ghee, Innovative techniques, Rapid test for adulteration.

Milk has been recognized as a complete food for millennia and persists to play an important role in the diets of over six billion people worldwide (Górska *et al.,* 2019; Indu, 2021). Fat, SNF, protein, lactose and ash are important milk constituents (Kumari *et al.,* 2018). Physico-chemical characteristics were used to assess changes in milk adulterated with various concentrations of adulterants such as urea, detergent, ammonium sulfate and neutralizers (Ahirwar *et al.,* 2015). Lipids are the main constituents of milk that play a major part in nutrition, aroma, physicochemical characteristics and financial aspects of the dairy sector (Patel, 2011). In Asian countries, particularly in India, milk fat is substantially consumed in the form of ghee (clarified butterfat) Ghee is considered to increase memory power, grasping power and strengthen the senses (Kaushik *et al.,* 2016; Ayari *et al.,* 2018). Ghee is the only fat containing short-chain fatty acids and is preferred over different fats generally due to being a significant source of fat-soluble vitamins and essential fatty acids, apart having unique aroma characteristics (Roy *et al.,* 2021).

Four methods such as direct cream method, the milk-butter method, the creamery-butter method and the pre-stratification method adopted to prepare ghee (Wadodkar *et al.,* 2002). The milk-butter (traditional) method is most preferable in India because, ghee prepared by this method have better than other industrial manufacturing processes but get a lesser cost (Wadodkar *et al.,* 2002). The process involves churning the fermented milk, which yields butter, called *makhan*. The butter is heated to 115°C to evaporate water. To obtain a golden yellow coloured ghee, which is strained using muslin cloth (Gandhi *et al.,* 2018) and stored at air tight container.

The ghee market is estimated to reach about 6.1 MT in 2020.The industry is projected to expand at a compound annual growth rate (CAGR) of 4.3% between 2021 and 2026, reach a volume of further 7.8 MT by 2026 (Global Ghee Market: By Source: Cow 2021).

Ghee adulteration is the process of purposeful mixing of inexpensive constituents of the same nature into pure ghee in proportions, to earn profit for dealers (Ayari *et al.,* 2018). Due to its high cost, ghee is vulnerable to adulteration with inexpensive fats, including animal fats, vegetable oil, hydrogenated fats, interesterified fats and inedible mineral oil (Gandhi *et al.* 2015; Gandhi *et al.* 2014; Kumar *et al.* 2013; Wasnik *et al.,* 2019). Fraudulent practise is a serious problem which must be assessed strictly to identify the adulterations in ghee (Roy *et al.,* 2021). The use of these affordable oil and fats is not only illegal, but poses significant

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How to cite this article: Kauser, H., Shilpashree, B.G., Praveen, A.R., Pushpa, B.P., Naik, H.D. and Ali, P. (2022). Innovation Techniques to Assess Adulterated Ghee: A Review. Bhartiya Krishi Anusandhan Patrika. 37(1): 71-75. DOI: 10.18805/BKAP452.

Submitted: 09-02-2022 **Accepted:** 09-05-2022 **Online:** 16-05-2022

health risks. Ghee contamination in India has been reported in electronic and print media on a regular base (Gandhi *et al.,* 2021). The methods available for the adulterations in ghee are based on chromatographic analysis (Rani *et al.,* 2015), spectroscopic analysis (Saleem,2020) thermal analysis (Upadhyay *et al.,* 2017) and physicochemical analysis (Gandhi *et al.,* 2018). These traditional technique are time-consuming, expensive and require severe sample processing before routine laboratory analysis (Roy *et al.,* 2021). Hence, innovation method are in trend to detect adulteration in ghee because of its speed, accuracy.

The current brief about various novel techniques to detect adulteration in ghee

ATR-FTIR and chemometrics

The attenuated total reflectance or ATR is, along with the transmission, one of the most commonly used sampling techniques during Fourier transform infrared spectroscopy. This technique is based on total internal reflection, where infrared (IR) light interacts with a sample only at the point of refraction. Meanwhile, transmission is based on the passing of IR light through the sample. An FTIR spectrometer is an extremely useful tool for determining the chemical composition of organic substances in solid, liquid, or gaseous form. Pre-treatment of samples is necessary for several sampling techniques to get good spectra. It is possible to measure solid or liquid samples with ATR FTIR spectroscopy with only minimal sample preparation (Griffiths *et al.,* 2007; Smith, 2011).

ATR-FTIR spectroscopy involves contact of the sample with the ATR crystal. The IR radiation travels through the crystal and interacts with the surface of the sample in contact with the ATR crystal. Due to the difference in refractive indices between the two materials, total internal reflection occurs. It is this reflection that forms the so-called "evanescent wave" that extends into the sample. Depending on the sample's composition, a small portion of the infrared light is absorbed when the evanescent wave interacts with the sample, resulting in an attenuated total reflection (Griffiths *et al.,* 2007, Smith, 2011).

FTIR (Fourier transform infrared spectroscopy) machines coupled with ATR (Attenuated total reflectance) are available from various vendors and quality control laboratories in the dairy industry. Method is rapid, non-destructive and low- cost, but knowledge of chemometrics is needed to completely exploit the eventuality of ATR-FTIR (Gandhi *et al.,* 2021).

The addition of coconut oil (2, 4, 6, 8, 10 and 15%) to ghee was detected using ATR-FTIR and chemometrics. The spectra of pure ghee, coconut oil (adulterated samples) were analysed in the wavenumber region of 4000-500 cm^{-1}. Principal component analysis (PCA) showed the distinct grouping of pure ghee and adulterated samples in the selected wavenumber range (1170-1141 and 1117-1100 cm^{-1}). Soft independent modelling by class analogy (SIMCA) was suitable to categorise the pure ghee and coconut oil samples of the confirmation set with a effectiveness of 100% using both partial least squares regression (PLS) and Principal components regression (PCR) models. The study revealed that coconut oil adulterated samples can be detected indeed at a lower concentration of 2% in ghee (Gandhi *et al.,* 2021).

Electronic nose system

An electronic nose (e-nose) based on metal oxide semiconductor (MOS) gas sensors was developed to detect adulteration in ghee with hydrogenated fat (vanaspati). To detect adulteration in ghee, data from the e-nose system were analysed for pattern recognition and classification Multivariate chemometric analysis methods, including principal component analysis (PCA) and discriminant function analysis (DFA). The PCA explained 98.10% of the variance in the e-nose dataset, while DFA explained 99.10%. The accuracy of training data and cross-validation was found to be 98.18% and 97.27%, respectively. Based on received e-nose signals, the DFA model was able to identify adulteration in 90.90% of the sample. The results of PCA and DFA suggest that the developed e-nose system successfully identified pure and adulterated ghee samples based on the e-nose data (Roy *et al.,* 2021).

Electronic nose devices have been designed to detect and distinguish various odours. Eight metal oxide semiconductor sensors were used to detect pure cow ghee from adulterated ones (mixed with sunflower oil and cow body fat), study conducted by Ayari *et al.,* 2018.

In this study, sunflower oil and cow body fat mixed with pure cow ghee (10%, 20%, 30%, 40% and 50%) were evaluated using a principal component analysis (PCA) and artificial neural networks (ANNs). Results show that the principal components analysis of sunflower oil and cow body fat were accurate to 96% and 97% of the variance, respectively. Based on the results, ANNs identified the adulteration with sunflower oil and cow body fat (Ayari *et al.,* 2018).

Complete liquefaction time

CLT (Complete liquefaction time) test the time it took for the solidified fat samples to melt completely at 45°C was measured and reported as complete liquefaction time (CLT) using the method (Kumar, 2008) with some changes. In brief, three grams of completely melted fat sample were placed in a test tube (length 10±0.1 cm, internal diameter 1.1±0.02 cm, external diameter 1.2±0.02 cm) and placed in a 60°C oven for 5 minutes. Following that, the test tube holding the fat sample was placed in a refrigerated (6-8°C) for 45 minutes to solidify. The solidified sample was then exposed to a liquefaction process at 45°C to finish melting. Using a stopwatch, the amount of time required for the sample to completely liquefy was recorded as CLT (Kamal *et al.,* 2018).

The addition of groundnut oil individually to cow and buffalo ghee was only detectable at a level of 15% adulteration using the CLT test at 44°C in cow ghee. However, animal body fat (Goat body fat) can be detected at 10 and 15% in buffalo ghee, but not in cow ghee. Detection

level of animal body fat in adulterated ghee varies from animal to animal (Upadhyay *et al.,* 2017). Pig body fat was detected at 15% levels in cow and buffalo ghee while buffalo body fat was detected at 10% levels (Kumar *et al.,* 2015). Based on a similar study conducted by goat body fat and groundnut oil combined in a ratio of 3:7 were detected in pooled cow and buffalo ghee above 20% and in cow ghee above 30% (Upadhyay *et al.,* 2017).

Normal-phase thin layer chromatography

Using normal phase thin layer chromatography (TLC), detect the presence of soybean oil and buffalo depot fat when mixed in cow and buffalo ghee at 5, 10, 15% (v/v). The samples unsaponifiable matter (USM) was extracted using diethyl ether after saponification of fats/oils with alcoholic potassium hydroxide and made alkali-free by washing with water, evaporation of the solvent and dissolving in chloroform. The unsaponifiable matter was spotted from all samples on TLC plates, along with some sterol reference standards (cholesterol, cholesterol acetate, phytosterols), β-carotene and tocopherols. The plates were prepared using solvent system (cyclohexane:ethyl.acetate:water:600:200:1,v/v). According to the results, soybean oil was easily detected in both types of ghee studied, even at a level of 5%, by obtaining three extra spots or bands (Rf values 0.465, 0.512, 0.557) in the reference area when compared to pure samples, whereas buffalo depot fat did not show any additional spots when compared to pure samples of cow and buffalo ghee (Kumar *et al.,* 2013).

Thin layer chromatography (TLC) of unsaponifiable matter the plates were developed by using solvent system (cyclohexane:ethyl.acetate:water:600:200:1, v/v) for normal-phase system Palm olein could be detected even at a lower 5% levels while sheep body fat could not be detected at any levels when added to ghee using Cholesterol and β-sitosterol (Gandhi, 2014).

Reversed-phase thin layer chromatographic (RP-TLC)

The minimal detection limit of milk adulteration using adulterant oils (rice bran oil, soybean oil, sunflower oil and groundnut oil) was determined using RP-18 plates to be 1%. Because the RP-TLC technique is simple and easy to execute in any laboratory testing milk quality and does not require any sophisticated instrumentation, it can be recommended to legal authorities such as FSSAI or BIS for inclusion in test methods (Upadhyay *et al.,* 2015).

RP-TLC is a simple, fast and accurate technology that has the potential to detect milk fat adulteration with inexpensive vegetable oils. Two solvent systems comprised of petroleum ether:acetonitrile:methanol:ethyl.acetate (1:1:2:1) and petroleumether:chloroform:acetonitrile:2 propanol:ethylacetate (1.5:0.5:5:3:1) was chosen to detect the presence of added vegetable oil (groundnut oil, soya bean oil and sunflower oil) in milk fat using Alumina backed Silica Gel 60 RP-18 F254s TLC plates against the reference standards of cholesterol, cholesterol ester, β-sitosterol, campesterol, stigmasterol, ergosterol, α and δ tocopherol. It was concluded that solvent system 1 can detect adulteration even at 1% level, whereas solvent system 2 can detect adulteration of all vegetable oils at 2% level, owing to the presence of β-sitosterol as an indicator, tocopherol and a few additional unidentified spots due to their occurrence in vegetable oils only. As a result, solvent system 1 has the potential to identify adulteration with vegetable oils in milk fat at a 1% level to verify authenticity. It may be done in a routine laboratory with minimal equipment and the process is reasonably simple to set up (Upadhyay *et al.,* 2015).

Using the reversed-phase thin layer chromatographic protocol standardised in the present study the adulteration of ghee with adulterant oils such as soybean oil, sunflower oil and groundnut oil could be detected up to 1% level while, designer oil up to 2% level. Similarly, coconut oil addition in ghee could be detected up to 7.5% level (Rani *et al.,* 2015). RP-TLC were developed by using suitable solvent system (petrol ether:acetonitrile:methanol:1:2:2v/v) palm olein could be easily detected even at 5% levels due to the presence of spot β-sitosterol, α-tocopherol, δ-tocopherol which is not there in case of pure sample of ghee (cow and buffalo) while sheep body fat could not be detected even up to a level of 15% using RP-TLC (Gandhi, 2014).

Chromogenic test

Adulteration of palm oil in ghee was identified by this method Palm oil was added to ghee at percentages of 5, 10, 15 and 20%. The detection limits in the range of 5%. The technique has proven to be simple and effective for detecting palm oil in ghee (Ramani *et al.,* 2019).

With the Potassium Ferricyanide and Ferric Chloride reagents, ghee samples adulterated with palm oil turned Prussian blue (Bector and sharma, 2002) Substances having reduction potential can react with potassium ferricyanide, which then reacts with ferric chloride to generate a ferric ferrous complex with an absorbance maximum of 700 nm (Jayanthi and Lalitha, 2011). Palm oil, as a result, contains carotenoids (alpha, beta and gamma carotenes), vitamin E (tocopherols and tocotrienols), sterols (sitosterol, stigmasterol and campesterol), phospholipids, glycolipids and squalene, all of which are effective water-soluble antioxidants (Neo *et al.,* 2008) when palm oil is added to ghee, the natural antioxidants in palm oil are employed to boost the colour of the chromogenic solution, which is commonly transformed to a deep blue colour. However, because pure ghee lacks natural antioxidants, the colour remained constant for a longer amount of time. It was also suggested that palm oil contain very little tannin, hence a Prussian blue colour was created due to the presence of palm oil in ghee using potassium ferricyanide and ferric chloride reagents (Ramani *et al.,* 2019).

DPPH (2, 2-diphenyl-1-picrylhydrazyl) based chromogenic assay been used to identify the presence of palm oil in ghee.

The interaction with DPPH solution resulted in the colour of pure ghee remaining violet, but the colour of ghee

adulterated with palm oil turning yellow. As a result, even with a 5% addition of palm oil to ghee, the colour changed pale yellow. It was also discovered that as the concentration of palm oil increased, so increase in the intensity of the yellow colour (Ramani *et al.,* 2018).

Image analysis

Detection of adulteration in cow ghee with vanaspati employing particle analysis and colour measurement, an image analysis technique. Image J, an accessible software and a flatbed scanner were used to process images and acquire images, respectively. Particle count, equivalent particle diameter, yellowness index and whiteness index were determined for pure cow ghee and compared to ghee contaminated with vanaspati at 5%, 10%, 15% and 20%. Adulteration can be detected using the particle count, equivalent particle diameter, brightness and whiteness index of cow ghee photographs. The created method is extremely cost-effective because it makes use of a low-cost flatbed scanner for picture capture, a readily available computer and free open-source Image J software. The suggested method can be utilized as an alternate strategy to identify vanaspati adulteration in cow ghee if it is adulterated at a level of 10% or higher (Wasnik *et al.,* 2019). The new approach can be automated for measurement, eliminating the need for manual procedures. The method is easily applicable as an alternative to routine laboratory adulteration detection for academic, research and quality control purposes. This image analysis approach has the advantages of convenience, quickness, accuracy, large sample size handling (difficult for manual detection), consistent results, no particular skills or training needs and was very cost-effective (Wasnik *et al.,* 2019).

Polymerase chain reaction (PCR)-based methodology

Develop a simple PCR-based method for detecting goat tallow in ghee. The kit-based isolation approach, which is effective for isolating a significant amount of DNA from a ghee sample, was utilized. The assay employs a goat species-specific primer, allowing for the detection of up to 10% levels of goat tallow adulteration in ghee and determining the type of body fat used to adulterate the milk fat (Hazra *et al.,* 2017).

CONCLUSION

The current situation has affected the reputation of the dairy industry both in India and abroad. The various incidences of adulteration reported in the literature used to assess the severity of the problem. Detecting of adulterants in ghee using a various quick, innovative approaches may bring awareness to maintain the standard quality of ghee. Implementation of their techniques in the scientific laboratories/referred labs could be source of income. Although, the intial investment is higher for certain techniques such as FTIR, electronic system but it is quick method to detect adulteration in ghee.

REFERENCES

Ahirwar, R., Harilal, P.T., Srihari, K.A. and Pandey, M.C. (2015). Quality changes in milk adulterated with detergent, urea, ammonium sulphate and neutralizers. Asian Journal of Dairy and Food Research. 34(4): 285-289.

Ayari, F., Mirzaee-Ghaleh, E., Rabbani, H. and Heidarbeigi, K. (2018). Detection of the adulteration in pure cow ghee by electronic nose method (case study: sunflower oil and cow body fat). International Journal of Food Properties. 21(1): 1670-1679.

Bector, B.S. and Sharma, V. (2002). Physico Chemical Characteristics of Ghee Adulterated with Palm Oil and its Detection. NDRI Annual Report: 44-45.

Gandhi, K. (2014). Detection of palm olein and sheep body fat adulteration in ghee using solvent fractionation technique. PhD. Thesis submitted to NDRI, Karnal, India.

Gandhi, K., Upadhyay, N., Aghav, A.D., Lal, D. and Sharma, V. (2014). Detection of palm olein and sheep body fat adulteration in ghee (clarified milk fat) using Reichert-Meissl (RM) value coupled with solvent fractionation technique. Indian Journal of Dairy Science. 67(5). DOI: 10.5146/IJDS. V67I5.40133.

Gandhi, K., Kumar, A. and Lal, D. (2015). Iodine value integrated with solvent fractionation technique as a tool for detecting palm olein and sheep body fat adulteration in ghee (clarified milk fat). Indian Journal Dairy Science. 68(4): 347-351.

Gandhi, K., Kumar, A. and Lal, D. (2018). Solvent fractionation technique paired with apparent solidification time (AST) test as a method to detect palm olein and sheep body fat in ghee (clarified milk fat). Indian Journal Dairy Science. 71(3): 246-251.

Gandhi, K., Sharma, R., Seth, R. and Mann, B. (2021). Detection of coconut oil in ghee using ATR-FTIR and chemometrics. Applied Food Research, (pp:100035). https://doi.org/10.1016/j.afres.2021.100035.

Griffiths, P.R. and De Haseth, J.A. (2007). Fourier Transform Infrared Spectrometry. John Wiley and Sons.

Global Ghee Market: By Source: Cow, Buffalo, mixed; by end use: Retail, institutional; by distribution channel: supermarkets/ hypermarkets, convenience stores, specialty stores, online, others; regional analysis; historical market and forecast (2017-2027); Market Dynamics: Competitive Landscape; Industry Events and Developments; https://www. expert marketresearch.com/reports/ghee-market. [accessed 2021 November 19].

Górska-Warsewicz, H., Rejman, K., Laskowski, W. and Czeczotko, M. (2019). Milk and dairy products and their nutritional contribution to the average polish diet. Nutrients. 11(8): 1771.

Hazra, T., Sharma, V., Sharma, R. and Arora, S. (2017). A species specific simplex polymerase chain reaction-based approach for detection of goat tallow in heat clarified milk fat (ghee). International Journal of Food Properties. 20(1). DOI: 10.1080/10942912.2017.1289542.

Indu, B. and Jayaprakasha, H.M. (2021). Conjugated linoleic acid-the natural trans fat: A review. Asian Journal of Dairy and Food Research. 40(4): 351-357.

Jayanthi, P. and Lalitha, P. (2011). Reducing power of the solvent extracts of *Eichhornia crassipes* (Mart.) solms. International Journal of Pharmacy and Pharmaceutical Sciences. 3: 126-128.

Kamal Gandhi, D., Rana, S. and Kumar, H. (2018). Solvent fractionation technique paired with complete liquefaction time (CLT) test to detect bland of palm olein and sheep body fat in ghee. International Journal of Comparative Sociology. 6(2): 458-463.

Kaushik, R., Jain, J. and Rai, P. (2016). Therapeutic potentials of cow derived products-A review. International Journal of Pharmaceutical Sciences and Research. 7(4): 1383. DOI: 10.13040/IJPSR.0975-8232.

Kumar, A., Sharma, V. and Lal, D. (2008). Development of a thin layer chromatography based method for the detection of rice bran oil as an adulterant in ghee. Indian Journal of Dairy Science. 61(2): 113-115.

Kumar, A., Neelam Upadhyay, K.G., Lal, D. and Sharma, V. (2013). Detection of soybean oil and buffalo depot fat in ghee using normal-phase thin layer chromatography. Indian Journal of Dairy Science. 66(4): 288-293.

Kumar, A., Upadhyay, N., Padghan, P.V., Gandhi, K., Lal, D. and Sharma, V. (2015). Detection of vegetable oil and animal depot fat adulteration in anhydrous milk fat (Ghee) using fatty acid composition. MOJ Food Processing and Technology. 1(3): 00013. DOI:10.15406/mojfpt.2015.01.00013.

Kumari, S., Pandey, A., Meena, P. and Choudhary, R. (2018). Milk protein and human health: A review. Bhartiya Krishi Anusandhan Patrika, 33(1/2):136-138.

Neo, Y.P., Ariffin, A., Tan, C.P. and Tan, Y.A. (2008). Determination of oil palm fruit phenolic compounds and their antioxidant activities using spectrophotometric methods. International Journal of Food Science and Technology. 43(10): 1832-1837.https://doi.org/10.1111/j.13652621.2008.01717.x.

Patel, A.M. (2011). Validation of methods for detection of ghee adulteration with animal body fat (Doctoral dissertation, NDRI).

Ramani, A., Hazra, T., Parmar, M.P., Sindhav, R.G. and Ramani, V.M. (2019). A simple rapid technique for detection of palm oil in ghee. Indian Journal Dairy Science. 72(4): 441-444.

Rani, A., Sharma, V., Arora, S., Lal, D. and Kumar, A. (2015). A rapid reversed-phase thin layer chromatographic protocol for detection of adulteration in ghee (clarified milk fat) with vegetable oils. Journal of Food Science and Technology. 52(4): 2434-2439. DOI: 10.1007/s13197-013-1208-3.

Roy, M., Manoj, D., Shanmugasundaram, S. and Yadav, B.K. (2021). Detection of adulteration in ghee (Clarified butter fat) using electronic nose combined with multivariate analysis.

Saleem, M. (2020). Fluorescence Spectroscopy Based Detection of Adulteration in Desi Ghee. Journal of Fluorescence. 30(1): 181-191. DOI: 10.1007/s10895-019-02483-0.

Smith, B.C. (2011). Fundamentals of Fourier Transform Infrared Spectroscopy. CRC Press.

Ramani, A., Hazra, T., Sudheendra, C.V., Hariyani, A.S., Prasad, S. and Ramani, V.M. (2018). Comparative appraisal of ghee and palm oil adulterated ghee on the basis of chromogenic test. International Journal of Current Microbiology and Applied Sciences. 7(12): 623-627.

Upadhyay, N., Kumar, A., Rathod, G., Goyal, A. and Lal, D. (2015). Development of a method employing reversed phase thin layer chromatography for establishing milk fat purity with respect to adulteration with vegetable oils. International Journal of Dairy Technology. 68(2): 207-217. DOI: 10.1111/1471-0307.12178.

Upadhyay, N., Goyal, A., Kumar, A. and Lal, D. (2017). Detection of adulteration by caprine body fat and mixtures of caprine body fat and groundnut oil in bovine and buffalo ghee using differential scanning calorimetry. International Journal of Dairy Technology. 70(2): 297-303.

Upadhyay, N., Kumar, A., Goyal, A. and Lal, D. (2017). Complete liquification time test coupled with solvent fractionation technique to detect adulteration of foreign fats in ghee. International Journal of Dairy Technology. 70(1): 110-118.

Wadodkar, U.R., Punjrath, J.S. and Shah, A.C. (2002). Evaluation of volatile compounds in different types of ghee using direct injection with gas chromatography-mass spectrometry. Journal of Dairy Research. 69(1): 163-171.

Wasnik, P.G., Menon, R.R., Sivaram, M., Nath, B.S., Balasubramanyam, B.V. and Manjunatha, M. (2019). Development of mathematical model for prediction of adulteration levels of cow ghee with vegetable fat using image analysis. Journal of Food Science and Technology. 56(4): 2320-2325.