

A simple rapid technique for detection of palm oil in ghee

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Abstract: The detection of palm oil adulteration in ghee continues to be a challenge for the dairy industry as well as food testing laboratories. Recently, various sophisticated instrument based protocols have been developed to detect a low level of vegetable oil in ghee but applications of those techniques in Indian condition is still a challenge, due to lack of modern instrument equipped food forensic lab facilities. Hence simple and rapid tests are very much essential to ascertain the quality of ghee. In the present investigation, a rapid chromogenic test based methodology has been standardized to detect palm oil in ghee. This standardized protocol did not show any false positive results in the genuine ghee samples. Adulteration of ghee with palm oil at the 5% level could be detected using the standardized chromogenic test. The protocol standardized is rapid and convenient to use.

Keywords: Adulteration Chromogenic test, Ghee, Palm oil

Food is the fuel of human civilization, so choice of food is directly concern to lifestyle, culture, religion and health for the modern civilization. Milk and dairy products has an integral relation with the human food culture. Milk is almost a complete food that provide almost all nutrients not only to the children but also old aged person. India is the leading milk producing country in the world and milk and dairy products plays equally an essential role

on the Indian economic growth. Not only fluid milk processing but also manufacturing of different milk products like ghee, butter, paneer and sweets are the key areas where organized and unorganized dairy entrepreneurs are getting major economical upliftment that directly involved in the growth of the country's economical structure. Milk lipid or fat that is a rich source of different bio active components, therefore it is supposed to be one of the costliest edible fat or one of the costliest component in dairy industry (Pereira et al. 2019). However, clarified milk fat or ghee is the second largest dairy product and almost 35% of the milk is converted into the ghee in Indian sub-continent (Hazra et al. 2017a). India produces 900,000 tonnes of marketed ghee, valued at Rs 85,000 million (Gandhi et al. 2018). As clarified milk fat; the ghee is a rich source of different bio active components including different fat soluble vitamins, essential fatty acids etc. The supremacy of ghee is not only acknowledged in Indian market only but also in developed country like USA (Wasnik et al. 2019).

Food adulteration is a an alarming threat to all over the world however the problem getting a serious dimension where there is a challenge between the physical supply and the market demand for a food item/items. Hence the food items like milk or milk products are always tense to be adulterated by unscrupulous traders for earning more money. Recently a report by FSSAI that 68% milk in India is substandard (Anonymous, 2018). The situation getting worst for premium quality of dairy products like ghee. The ghee production level varies from region to region, 57% in northern region, 9.5% in eastern, 23.5% in western and 10% in southern region (Aneja et al. 2002). The integral relation of ghee with Indian culinary as well as religion culture and uneven production of ghee; trends this premium product to be adulterated with cheaper quality of oils (vegetable oil) and fats. Recent scientific report stated that problem of ghee adulteration getting a serious concern for food law enforcement agencies, as crooks involved in the business of ghee production are using designed oils/ fats with or such oil that has almost same fatty acids profile of milk fat or ghee, so that BR reading at 40°C, RM value and Polenske value could be easily manipulated (Sharma and Pathania, 2019). Previously different techniques been reported based on image analysis (Wasnik et al. 2019 and Wasnik et al. 2017) Triglyceride analysis (Hazra et al. 2018 and Sharma et al. 2017),

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Infrared spectrum analysis (Pereira et al. 2019 and Aparnathi et al. 2019), olfactory component analysis using electronic tongue (Ayari et al. 2018), refractive index analysis using sensor (Annasamy et al. 2019) and DNA analysis using PCR (Hazra et al. 2017b); all these reported techniques are very fruitful and able to detect very low level of externally added fats or oils in ghee. However in Indian condition the modern sophisticated instrument equipped food forensic laboratories are very limited, so those sophisticated analysis are not easily available in routine quality control analysis. Recent studies reported that detection of palm oil in ghee or clarified milk fat is always a challenging task (Aparnathi et al. 2019). Earlier researchers were unable to detect palm oil adulterating in ghee less than 10% level (Lal et al. 1998, Gandhi 2014 and Aparnathi et al. 2019). Due to similar fatty acid profile of palm oil with milk fat, the detection of palm oil in ghee is always a challenging task indeed.

Considering all the facts reported here, we have tried to develop a rapid chromogenic test for detection of palm oil adulteration in ghee.

Mix milk was collected from local dairy farmer of Amreli district Gujarat. The ghee was prepared by creamery butter method. The final temperature of clarification was 110°C. The clarified fat was filtered using coarse filter paper to obtain ghee and stored in airtight glass bottles at room temperature.

Palm oil was procured from local market of Amreli city. For the preparation of adulterated ghee samples, pure ghee samples was heated to 60-70°C for 10 min before adding and mixing of palm oil. The adulterated ghee samples were prepared by addition of Palm oil in four different proportions (5%, 10%, 15% and 20% w/w) into pure ghee.

Accurately 1.297 gm. of Ferric Chloride (molecular weight: 162.20) weighed and dissolved in distilled water. Final volume was make up to 1000 ml in a volumetric flask using distilled water.

Accurately 11.04 gm. of Potassium Fericyanide (molecular weight: 368) weighed accurately and dissolved in distilled water. Final

volume was make up to 1000 ml in a volumetric flask using distilled water.

Two ml of milk fat/ghee was taken in a clean dry test tube. There after 1 ml of Ferric Chloride solution (0.008 M) was added in that test tube and there after 0.3 ml Potassium Fericyanide solution (0.03 M) was added and mix it for 30 seconds; thereafter the colour change was observed. For pure ghee the colour would be green and for adulterated ghee the colour would be changed green to blue.

Ferric Chloride of concentration (0.008 M) and Potassium Fericyanide (0.03 M) was used for present studies. However to standardize the said test protocol in initial phase of trials 1 and 2 ml fat solution were used respectively. The observation for these two said condition were given in table-1.

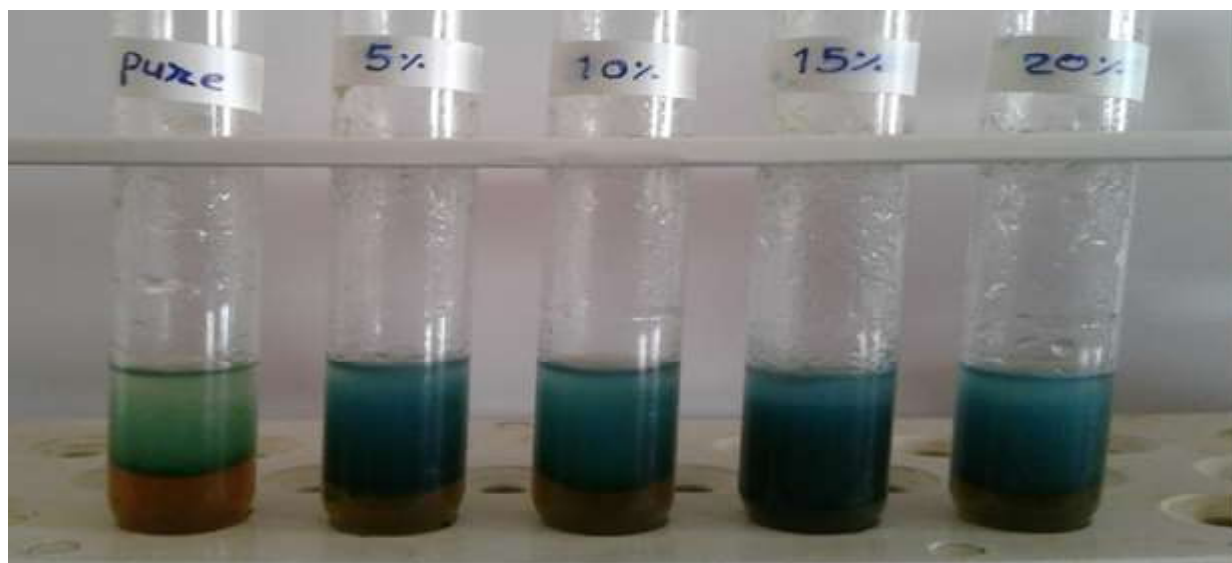
When 1 ml of pure ghee or adulterated ghee (5, 10, 15 and 20 percent level of adulteration) samples were allowed to react with the said chromogenic solutions; it was observed that green colour persisted in pure ghee beyond 2 minutes after the addition of chromogenic solution. However ghee samples added with 10, 15 and 20 percent level of palm oil adulteration; the colour turned from green to deep blue colour (purssian blue) after 1 minute of the addition of the chromogenic solutions. However, for ghee sample adulterated with 5% palm oil, the green colour remain persisted for more than 2 min; hence no color difference was observed for pure ghee and ghee added with 5% palm oil even after two minutes of the conductance of the test. So this said condition was eluded for further trials.

When 2 ml of pure ghee or adulterated ghee (5, 10, 15 and 20 percent level of adulteration) samples were allowed to react with the said chromogenic solution; it was observed that green colour persisted in pure ghee beyond 2 minutes after the addition of chromogenic solution; while ghee samples with 5, 10, 15 and 20 percent level of palm oil adulteration; the colour turned from green to deep blue colour (purssian blue) after 1 minute of the addition of the chromogenic solution. Hence clear colour

Table 1 Time taken for the reaction between ghee samples with Ferric chloride and Potassium Fericyanide

Samples	Quantity of Ghee	Time taken	Observation
Pure Ghee	1 ml	More than 2 min.	Green colourPersists
Pure Ghee + 5% PO*	1 ml	More than 2 min.	Green colourPersists
Pure Ghee + 10% PO	1 ml	More than 1 min.	Green to dark green colour
Pure Ghee + 15% PO	1 ml	More than 1 min.	Dark green colour
Pure Ghee + 20% PO	1 ml	1 min.	Dark green colour
Pure Ghee	2 ml	More than 1 min.	Light green colourPersists
Pure Ghee + 5% PO	2 ml	More than 30 sec.	Lime green colour
Pure Ghee + 10% PO	2 ml	More than 30 sec.	Lime green to green colour
Pure Ghee + 15% PO	2 ml	30 sec.	Green colour
Pure Ghee + 20% PO	2 ml	30 sec.	Dark green colour

Note: *PO – Palm oil



(Pure Ghee, Pure ghee with 5% palm oil, pure ghee with 10% palm oil, pure ghee with 15% palm oil, pure ghee with 20% palm oil.)

Fig. 1 Reaction between ghee samples, Ferric chloride and Potassium Ferricyanide after 5min of conductance of test

difference was observed between pure ghee and ghee adulterated with even 5% palm oil.

From figure (1) it was observed that significant colour difference was observed between pure ghee and adulterated ghee. Hence this said protocol condition able to detect even 5% level palm oil adulteration in ghee.

Ghee samples adulterated with palm oil turned to prussian blue colour with Potassium Ferricyanide and Ferric Chloride reagent (Bector and Sharma 2002). Substances that have reduction potential able to react with Potassium Ferricyanide then reacts with Ferric Chloride to form ferric ferrous complex of intense blue color that has an absorption maximum at 700 nm (Jayanthi and Lalitha, 2011). Natural antioxidants having potential reduction effect, hence the palm oil contains carotenoids (alpha, beta and gamma carotenes), vitamin E (tocophoreols and tocotrienols), sterols (sitosterol, stigmasterol and campesterol), phospholipids, glycolipids and squalene, which are powerful water-soluble antioxidants (Yun et al. 2008). Whenever palm oil added in ghee; the natural antioxidants those are present in palm oil use to be increased so the colour of the chromogenic solution used to be changed to deep blue colour. However due to dearth of natural antioxidants in pure ghee the colour remained stable for longer period of time. It was also recommended that palm oil contain a very small amount of tannin so prussian blue colour with potassium ferricyanide and ferric chloride reagent formed due to presence of palm oil in ghee.

Recent studies reported that detection of palm oil in ghee is still a challenge (Aparnathi et al. 2019). FSSAI recently recommended unsaponifiable component analysis, for detection of vegetable oil in ghee using HPLC. (Anonymous, 2019). However this said

protocol not able to detect palm oil in ghee. So far in this situation this simple test could be an effective alternative solution for detection of palm oil adulteration in ghee. For checking the repeatability of this said test, this said protocol was used fifty times and same results was observed in each and every trial. Similar type of results were observed for ghee samples adulterated with other vegetable oils like cotton, ground nut and soy oil.

Conclusions

A chromogenic analytical method was developed for detection of palm oil adulteration in ghee. Palm oil was added in ghee @ 5, 10, 15 & 20 %. Limits of detection was to the tune of 5%. The method proposed proved to be simple and appropriate for the detection of palm oil in ghee.

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