CHAPTER VII
RHEOLOGICAL PROPERTIES AND SENSORY CHARACTERISTICS OF GREEN TEA YOGURT DURING STORAGE

7.1 Introduction

Yogurt and related fermented dairy products have considerable economic importance worldwide owing to their high nutritional values. For instance, most dairy products usually contain substantial amount of highly bioactive compounds produced as a result of enzymes breakdown of milk proteins (Allen, 1982). This strategic property can be further diversified by adding nutraceutical (nutritional and pharmaceutical) ingredients such as plant extracts rich in phytochemicals to the yogurt (Guggisberg, Eberhard, & Albrecht, 2007). In the present study green tea rich in polyphenolic compounds was used to fortify yogurt, thus making herbal-yogurt a new fermented food category targeting consumers with variety health issues associated with beneficial effects of green tea consumption (Narotzki, Reznick, Aizenbud, & Levy, 2012). However, the presence of green tea affected the fermentation of milk resulting in increased acidification and proteolysis (Section 5.3.1 and 5.3.7 respectively.). These are expected to alter the coagulation of milk protein and subsequently the modification of textural and rheological properties of yogurt. Determining
the consistency of the yogurt gel network by rheological methods is a challenge because the structure can be partly damaged in the act of sample preparation. In addition factors related to yogurt formation i.e a) the heat treatment/homogenization of the milk base (casein/whey protein ratio), b) the starter culture, and c) technological influences such as temperature, pressure, valves and the shear history are also known to be an important determinant of yogurt structure (Sodini, Remeuf, & Haddad, 2004). The starter culture in turn may also be influenced by the presence of green tea. The objectives of the present study were to determine the influence of green tea supplemented yogurt on its sensory characteristics and various physical and rheological properties, WHC, whey separation and total solid values during storage.

7.2 Materials and Methods

Samples preparation and methods for experiments carried out in this section were as described in Sections 3.12, 3.13, 3.14 and 3.15.

7.3 Results and Discussion

7.3.1 Effects of green tea on rheological characteristics of refrigerated yogurt

For rheological measurements yogurts were stirred, as consumers usually do before consumption, prior to sampling. This is the most used method to perform viscometry or oscillation tests on yogurt probably because it is difficult to find and standardize a mechanical and reproducible method to stir yogurts without breaking substantial amount of their structure (Vercet, Oria, Marquina, Crelier, & Lopez-Buesa, 2002). Manual slow stirring is regarded as the most effective way to preserve yogurt structure and at the same time allow preparation of yogurt sample that can be measured in a rheometer equipped with
parallel plate geometry (Vercet et al., 2002), suggested that when the yogurt network is very weak, sedimentation of casein aggregates occur, and that the syneresis triggered by this agglomeration leads to the formation of a depleted layer at the upper surface of the sample. When oscillatory measures are to be taken in a horizontal geometry, this layer causes slippage of the moving element, thus giving rise to an apparent reduction in modulus.

7.3.1.a Apparent viscosity

The apparent viscosity of plain and green tea yogurts is shown in Figure 7.1. All three yogurts showed shear thinning properties, demonstrated as decreased viscosity as the rate of shearing increased (at higher shear rate the viscosity of the yogurt decreases making it more runny). If time is considered as variable factor, the fluid may show rheopectic behavior. This is generally due to a reversible change in the structure of the material with time under shear, with a limiting viscosity ultimately being approached (Velez-Ruiz & Barbosa Canovas, 1997).

Green tea yogurts showed a significant decrease in viscosity with time as compared to plain yogurt at all storage periods studied. On the 1st day, the initial apparent viscosity of plain yogurt (291.3 Pas) was higher than MGT and JGT-yogurts (86.41 and 76.98 Pas; p<0.05; Figure 7.1). Although refrigerated storage had significant (p<0.05) effect on initial apparent viscosity for green tea-yogurts (86.41-107.8, 76.98-115.7 Pas for MGT and JGT-yogurts respectively; Figure 7.2, 7.3 and 7.4), but caused adverse effects on viscosity in plain yogurt as demonstrated by the reduced viscosity on day 28 (230.8 ±1. 7 Pas) compared to that in fresh yogurt (291.3 ±1. 64 Pas). Decreasing viscosity during storage is not beneficial for industrial market and consumers acceptance.
Figure 7.1 Viscosities of fresh plain, Malaysia green tea and Japanese green tea-yogurt
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Figure 7.2 Viscosity for plain yogurt during storage

Figure 7.3 Viscosity of Malaysian green tea yogurt during storage
Figure 7.4 Viscosity of Japanese green tea yogurt during storage

7.3.2 Dynamic rheological

7.3.2.a Frequency sweep

Yogurt is a viscoelastic material, so that its rheological behavior can be described by two parameters; namely, the storage (G’; elasticity) and the loss (G”, viscosity) module (Penna, Sivieri, & Oliveira, 2001; Ramirez-Santiago et al., 2010).

G’ is a measure of the energy stored and subsequently released per cycle of deformation per unit volume. It is the property that relates to the molecular events of an elastic nature. G” is a measure of the energy dissipated as heat per cycle of deformation per unit volume. G” is the property that relates to the molecular events of viscous nature. Another commonly used dynamic viscoelastic property, the loss tangent (which equals G”/G’) denotes relative effects of viscous and elastic components in a viscoelastic behavior.

Figure 7.5, 7.6, 7.7 and 7.8 shows the viscoelastic behavior of the fresh and refrigerated plain- and green tea –yogurts during storage, which occurs in practice when samples are taken out of the refrigerator for consumption and then stored again. Elastic modulus dominating viscous modulus (G’>G”) showed that yogurt is having solid behavior for all of the three yogurts type. As the frequency increases the G’ and G” increases gradually. These results are in accordance to Sendra et al., (2010) which showed yogurt’s predominantly elastic behavior (G’ > G”) over the whole range of frequencies tested (0.01-10 rad/s) and which corresponds closely to that of a true gel. G’ higher than G” can be due to decreased electrostatic repulsion and increased casein-casein interactions (Rao, 2003). Fresh plain yogurt showed a higher range of G” and G’ than the green tea-yogurts (31.34- 72.84, 12.73- 37.26 and 11.29 - 36.23 Pa, for PY , MGTY and JGTY
respectively; p<0.05; Figure 7.5), which is showing the improvement of viscoelasticity of the sample due to higher solid content.

There was no significant (p>0.05) difference in the Elastic modulus and viscous modulus of plain and green tea yogurts during of storage. This indicates that the no significant change in the bonding involved in the formation of gel structure by adding green tea, which may be attributed to the function of phenolic compounds in green tea that no interacted as a cross-link between protein molecules. Therefore, in green tea yogurt, the nature of bonding did not change but the extent of cross-linking increased due to the presence of dissolved phenolic compounds.

However, on 21\textsuperscript{th} day and 28\textsuperscript{th} day of storage, plain yogurt showed statistically different $G''$ and $G'$ from that on 1\textsuperscript{th} day, 7\textsuperscript{th} and 14\textsuperscript{th} day; however, the difference was very small (p>0.05). This shows an increase in solid like characteristics of the samples on storage for 2 weeks which may be due to increase in the acidity of the samples during storage.
Figure 7.5 Frequency sweep of fresh plain yogurt, Malaysia green tea yogurt and Japanese green tea yogurt.

Figure 7.6 Frequency sweep for Malaysia’s green tea yogurt during storage.
Figure 7.7 Frequency sweep for Japanese green tea yogurt during storage
7.3.2.b Amplitude sweep

Amplitude oscillatory rheology has been used to characterize the rheological properties of yogurt during the gel formation process (fermentation) without damaging the weak gel network. Small deformation is defined as a small relative deformation (strain or change in dimension) (e.g. \( \leq 1\% \)), which when applied does not disrupt the development of the network structure, i.e., within the linear viscoelastic region. In this “linear” region, the dynamic moduli are independent of the applied stress or strain (Lee & Lucey, 2010).

Figure 7.9 shows the values of amplitude sweep for fresh plain and green tea-yogurts. The critical strain of fresh plain yogurt was the highest (44.07 - 16.58 Pa) followed by JGTY and MGTY (27.9 - 6.95 and 24.24 - 7.69 Pa respectively) in range of 0.0004 - 0.10 strain. The elevated critical strains (yield point) suggest that the plain yogurt had a higher resistance to deformation. The high elastic modulus (\( G' \)) for plain yogurt indicates it...
has more solid like and the binding particles (in this case milk protein) is packed closer than those in green tea-yogurts.

While JGTY showed lower elastic modulus ($G'$) with initial strain (0.0004) compared to plain yogurt suggested that it had lower stiffness and weaker than plain yogurts. It had a slightly higher critical strain than Malaysia green tea yogurt hence it is moderate in nature, followed by MGTY which had the lowest elastic modulus ($G'$) with same strain compared with plain yogurt, suggested that it has less solid behavior (Figure 7.9). The differences in the storage modulus reflected the gelation characteristics within the different yogurts.

Figure 7.10, 7.11 and 7.12 showed amplitude sweep of samples during storage. PY showed the highest value of elastic modulus ($G'$) in 7\textsuperscript{th} day of storage ($66.68$ - $28.73$ Pa; Figure 7.10) with the lowest value on the 28\textsuperscript{th} day of storage ($38.76$ - $15.59$ Pa). Suggest that in 7 day yogurt was more solid like and the particles (proteins) are closely packed. Present of green tea to yogurt indicated no effect on yogurts during storage. The changes in values of elastic modulus ($G'$) for both green tea yogurts were not significant .Except for JGTY in 28\textsuperscript{th} day of storage which showed highest elastic modulus value ($56.44$ - $23.56$ Pa; Figure 7.12). The lower value observed in yogurts fortified with the green tea can be attributed to the production of exopolysaccharide by the green tea. The filaments of exopolysaccharides interfere with the casein network. It can be assumed that protein strand formation and protein–protein interaction is partly inhibited by the exopolysaccharides, thus reducing the rigidity of the resulting yogurt gel.
Figure 7.9 Amplitude sweep of fresh plain, Malaysia green tea and Japanese green tea- yogurts

Figure 7.10 Amplitude sweep for plain yogurt during storage
Figure 7.11 Amplitude sweep for Malaysian green tea yogurt during storage

Figure 7.12 Amplitude sweep for Japanese green tea yogurt during storage.
7.3.3 Effects of green tea on sensory evaluation of yogurt

The effects of adding green tea on sensory properties of yogurt samples are shown in Figure 7.13. Yogurt tasters play an important role in the quality assessment of samples. Yogurt tasters’ parameters for evaluation included appearance, flavor, color, texture and aroma.

The presence of green tea in yogurt increased yogurt appearance (7.1±1.5 and 6.6±1.3 for MGT and JGT–yogurts respectively) compared to plain-yogurt (5.1±1.3), indicating that both MGT and JGT yogurts were preferred by the panellists.

Texture score were lower for MGT and JGT–yogurts (6.4±1.7 and 6.1±1.9 respectively) compared to plain (7.4±1.7). Plain yogurt on the other hand showed low flavor score (5.9±1.5) compared to MGT and JGT–yogurts (7.45±1.5 and 7.5±1.4 respectively). Colour has much impact on the acceptability of the green tea yogurt samples (8.48±1.8, 8.0±1.7 and 5.9±2.2 for MGTY, JGTY and PY respectively). Green tea also increased (p<0.05) the fresh yogurt score for aroma (8.9±1.3 and 8.3±1.5 for MGTY and JGTY respectively) and the overall score (8.9±1.8 and 8.9±0.8 for MGTY and JGTY respectively) compared to plain-yogurt (5.3±2.3 and 6.5±1.8 respectively).

In recent years, per capita consumption of yogurt has increased drastically because many consumers associate yogurt with good health (Karaolis, Botsaris, Pantelides, & Tsaltas, 2013). Yogurt is characterized as a fermented milk product with a refreshing flavor, a smooth viscous gel, and a slight sour taste (Bodyfelt, Tobias, & Trout, 1988). These sensory properties in addition to appearance, flavour, texture, and overall quality offer quality control criteria (Isleten & Karagul-Yuceer, 2006) and thus were used in the evaluation of yogurt. The mean sensory characters scores for all samples were 5.0 or higher (Table 7.1). The hedonic score of 5 corresponds to moderate liking of the samples, and the hedonic
score of 8 corresponds to very much liking of the samples. Both green tea yogurts were liked moderately by the consumers for their texture, which were less than considered for plain yogurt.

Since the flavour of yogurt is affected by the rate of acid production during the fermentation process (Gallardo-Escamilla, Kelly, & Delahunty, 2005), the high score of flavour in both green tea yogurts indicate that the addition of green tea contribute to more acid production from conversion of lactose to lactic acid. This finding is important because the addition of some lactic acid bacteria can affect flavour compounds.

Sensory evaluation showed that the plain (5.3±2.3) and the green tea yogurts (8.9±1.3 and 8.3±1.5 for MGTY and JGTY respectively) scored significantly higher (P < 0.05) aroma measurement. Acetaldehyde was firstly reported by Pette & Lolkema (1950) as the main aromatic compound in yogurt. During manufacture, production of this compound is only highlighted when a certain level of acidification is reached (pH 5.0). The maximum amount is obtained at pH 4.2 and stabilizes at pH 4.0. The production of acetaldehyde and other flavour compounds by S. thermophilus and Lb.bulgariicus occurs during yogurt fermentation and the final amount is dependent on specific enzymes which are able to catalyse the formation of carbon compounds from the various milk constituents. This result was consistent with changes in pH and titratable acidity values that is, yogurts with high acidity contained the highest amount of acetaldehyde. Since the present of green tea to milk increased acidity of yogurts (see Table 5.1 and 5.2) it was suggested that decomposition of aroma compounds occurred more in green tea yogurts than plain yogurt during fermentation.

Overall, significant difference was observed between green tea yogurts (8.9±1.8 and 8.4±0.8 for MGTY and JGTY respectively) and plain yogurt (6.5±1.8) in the scores of
overall acceptability; even though no significant difference was observed for MGTY and the JGTY samples. This indicated that addition of green tea affected the overall acceptability of yogurt due to the higher score of colour, aroma as well as the acceptable flavour and appearance.

Green tea yogurt has the potential to attract new yogurt consumers because the incorporation of green tea into yogurt can enhance the therapeutic value of yogurt (see Section 6.3.8). Green tea yogurt could provide novelty in the dairy foods market and help consumers obtain nutritious food with added health benefits.

Figure 7.13 Sensory analysis results of plain and green tea yogurts.

7.3.4 Effects of green tea on water holding capacity (WHC) of yogurt during storage

Water holding capacity (WHC) is related to the ability of the proteins to retain water
within the yogurt structure (Wu, Hulbert, & Mount, 2001). The mobility of water molecules in yogurt, as reflected in WHC values, can affect yield, sensory evaluation, stability (in physical terms) and texture. In fact WHC is an essential quality parameter such that the viscosity can be increased, gel-structures can be created or the physical stability can be lengthened by changing the WHC (Mao, Tang, & Swanson, 2001). Thus changes in WHC as a result of functional additive such as herb water extracts may modify the properties of yogurt in a predetermined manner.

In this study, WHC measurements showed significant differences between green tea and plain yogurt samples (Figure 7.14). The higher WHC was obtained for green tea yogurt samples (30.2±1.2 and 31.25± 0.67 % for MGTY and JGTY respectively p<0.05) than those obtained in plain yogurt (26.05± 1.5 %) in first day which was 2.92 and 3.02% higher than that of plain yogurt. The WHC of plain and green tea yogurts increased during first week (34.4±1.6, 33.8±1.1 and 27.71±0.46% for MGTY, JGTY and PY respectively on 7th day of storage) and thereafter it decreased (27.48±1.75, 27.75±2.2 and 23.98±0.96 % for MGTY, JGTY and PY respectively on 28th day of storage). Decrease in WHC of yogurt during storage was observed by Sahan, Yasar, & Hayaloglu (2008). Decrease WHC in yogurts during storage is partly due to the unstable gel network of yogurts, in which the weak colloidal linkage of protein micelles cannot entrap water within its three-dimensional network (Donkor, Nilmini, Stolic, Vasiljevic, & Shah, 2007). However Parnell-Clunies, Kakuda, Mullen, Arnott, & DeMan (1986) hypothesized that as the β-lactoglobulin interacted with κ-casein, more covalent bonds were formed and larger micelle sizes might cause steric hindrances; all resulting in lower WHC (covalent bonds decrease the number of charged groups present in the gel network) during storage.
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In yogurts, increased micelle size and increased whey-casein and casein-casein interactions lead to a more porous gel, which could retain more water (Sodini, Montella, & Tong, 2005; Lee & Lucey, 2004).

Figure 7.14 Water holding capacity of yogurt by adding green tea during storage. Values are mean±SD. (n=3).

7.3.5 Effects of green tea on changes of syneresis on yogurt during storage

Syneresis is defined as gel contraction that occurs concomitantly with liquid/whey expulsion and relates to the inability of the gel network to entrap all of the liquid phase. Most consumers consider syneresis to be a defect (Lucey, Tamehana, Singh, & Munro,
When the casein particles rearrange in the gel network, whey expulsion is spontaneous, as the gel shrinks without the application of some external force (Lucey, 2002).

The syneresis (%) of all 3 types of fresh and stored yogurt is given in Figure 7.15. On the first day of storage the green tea yogurts showed the lower syneresis (3.29±0.89 and 3.26±0.97 %) than those in plain yogurt (3.56±1.1 %; p>0.05). All yogurts showed an increase in the amount of syneresis up to 28 days of storage (4.13±0.3, 4.18±1.0 and 4.91±1.3%; for MGTY, JGTY and PY respectively; p<0.05) at 4 °C. These findings are in agreement with those of Ramirez-Santiago et al. (2010); Zare, Boye, Orsat, Champagne, & Simpson (2011) who showed an increase in the extent of syneresis in yogurts with refrigerated storage. An increase in syneresis with storage time was observed in all yogurts. Although the phenomena occurring during syneresis are not fully understood, it is agreed that increased syneresis with storage time is usually associated with severe casein network rearrangements (Van Vliet, Lucey, Grolle, & Walstra, 1997; Ramirez-Santiago et al., 2010), that promote whey expulsion. Functionality of hydrocolloids in yogurt is demonstrated by their ability to bind water, interact with the milk constituents (mainly proteins), and stabilize the protein network, preventing free movement of water (Tamime & Robinson, 1999).

Whey separation is known to be related to instability of gel network and thus the loss in ability to entrap all the serum phase (Lucey, 2002). The rate of acidification is instrumental in whey separation of yogurt in which the faster the rate the poorer the network rearrangement during whey expulsion thereby resulting in lesser whey separation (Castillo et al., 2006). The shorter time for green tea yogurts than plain yogurts to reach pH
4.5 (see section 5.3.1) may result in the reduction in whey separation in MGTY and JGTY as compared to plain-yogurt.

![Figure 7.15 Effects of Green tea extract on the syneresis of yogurt](image)

Figure 7.15 Effects of Green tea extract on the syneresis of yogurt

7.3.6 Effect of green tea on total solid content in yogurt

The total solid (TS) content of yogurt samples showed in Figure 7.16. It was observed from the results that the total solids content in the MGTY was in the range of 9-13%, and JGTY was in the range of 8-13% while PY was in the range of 7-13%.

These TS values were much lower than those in commercial yogurts which have a typical range of 14-15% (Tamime & Robinson, 1999). This is because yogurt prepared in the present studies was without the addition of stabilizers or milk powder which commonly practiced in the commercial preparation of set or stirred yogurt. The apparent decrease in total solids with storage time (8.72, 8.44 and 6.97% for MGTY, JGTY and PY
respectively) in day 28 may be explained by moisture loss which had an advantage of increased syneresis (Figure 7.14). The total solids content of the yogurt samples had a significant effect on the degree of syneresis. Reduction of free water and increasing the proportion of solids content, which occur during fermentation, are two main factors decreased rates of wheying off in the samples with high total solids. This is agreeing with the finding by Jaros, Partschefeld, Henle, & Rohm (2006); Amatayakul, Sherkat, & Shah (2006); Mahdian & Tehrani (2007) who found that increase whey separation of yogurt occurred only when the total solids were decreased. In this regard the addition of green tea may be seen as advantageous in reducing the wheying off while yogurt is being kept refrigerated.

![Figure 7.16 Changes in total solid content of yogurts during refrigerated storage](image)

Figure. 7.16 Changes in total solid content of yogurts during refrigerated storage
7.4 Conclusion

The addition of green tea (2 g), as a source of phytochemicals, to supplement yogurts appear to decrease viscosity behavior, elastic modulus and amplitude sweep than those found in plain yogurt but promoted avenue for increased sensory properties, with high consumer acceptability. Increased WHC in green tea yogurts contributed to lower syneresis and higher total solid content. Both green teas itself are well known for their beneficial health effects, and together with yogurt they may constitute a functional food with commercial applications.
References


