



A Study of the Effect of Probiotics on Antioxidant Activities of Phenolic Compounds in Functional Yoghurt Fortified with Fruit

By

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SUMMARY

Dairy products are suitable for functional meals since they have a high nutritional value. Yogurt is a fermented dairy product obtained from milk by lactic acid fermentation. Yogurt is remarkable with its nutritional value over the other fermented dairy products. Fermented milk products help to maintain overall gastrointestinal health. Prebiotics are food components that are either non-digestible or low-digestible because they encourage the growth of certain probiotic bacteria in the colon. Prebiotics are exceptional tools to promote the growth of beneficial gut microorganisms that ferment them and produce short chain fatty acids (SCFAs) and vitamins.

Fermented milk with probiotic microorganisms provides a number of health advantages, including lowering blood cholesterol, reducing diarrhea, preventing and suppressing colon cancer, and boosting the immune system.

Fruits are abundant sources of dietary fiber, vitamins, and minerals, as well as carbohydrates. Plum (*Prunus*

domestica L.) is rich in phenolic compounds, anthocyanins, carotenoids, minerals, vitamins (A, B, C, and K), and pectin, among other bioactive substances.

Mango (*Mangifera indica* L.) is a fantastic source of dietary fiber, which is crucial for human nutrition and health, as well as bioactive substances such pro-vitamin A, carotenoids, vitamin C, and phenolics.

Pomegranate (*Punica granatum* L.), is content bioactive substances such phenolics and flavonoids, primarily anthocyanins. It contains phytochemicals, vitamins, and minerals, and macronutrients. Pomegranate juice has protective effects against atherosclerosis, lowdensity lipoprotein oxidation, prostate cancer, platelet aggregation and cardiovascular disorders.

The aim of this study is to investigate the effect of probiotic on antioxidant activity of phenolic compounds in functional yogurt fortified with plum, mango and pomegranate juices. On the other hand, the physiochemical, rheological and microbial properties of probiotic yogurt fortified with different fruit juices were determined. In addition to study the acceptability of probiotic yogurt fortified with different fruit juices.

Yogurt manufacturing was started with heating the milk at 95 °C for 5 min, and then it was cooled to 42 °C. After that 10 % of pasteurized plum, mango and pomegranate juice were added and divided into the following eight formulas as follows:

1st Formula (Control yogurt): Milk was inoculated with 2 % of yogurt culture, which contain *L. bulgaricus* and *S. thermophilus culture* (1:1v/v). 2nd Formula (Probiotic yogurt): Milk was inoculated with 2 % of yogurt culture, and *Bifidobacterium infantis* culture .3rd Formula: Yogurt fortified with 10% plum juice. 4th Formula: Yogurt fortified with 10% mango juice. 5th Formula: Yogurt fortified with 10% pomegranate juice. 6th Formula: Probiotic yogurt fortified with 10% plum juice. 7th Formula: Probiotic yogurt fortified with 10% mango juice. 8th Formula: Probiotic yogurt fortified with 10% mango juice. 8th Formula: Probiotic yogurt fortified with 10% pomegranate juice.

1-Physicochemical analysis of different juice samples plum, mango and pomegranate and different yogurt samples

The chemical composition of plum, mango and pomegranate juices were $(18.67\pm0.01, 30.2\pm0.02)$ and $21.9\pm0.03\%$ for total solid, $(80.44\pm0.01, 68.8\pm0.02)$ and $78.1\pm0.03\%$ moisture, $(0.04\pm0.01, 0.07\pm0.01)$ and

 $0.32\pm0.02\%$) fat, $(0.62\pm0.01,\ 0.11\pm0.01\ \text{and}\ 1.31\pm0.02\%)$ protein and $(0.23\pm0.01,\ 0.20\pm0.02,\ \text{and}\ 0.37\pm0.02\%)$ ash respectively. While the mean values of total phenol, total flavonoid and antioxidant activities of plum, mango and pomegranate juices were $(108.782\pm0.21,\ 38.055\pm0.19\ \text{and}\ 103.69\pm0.22\ \text{mg/}100\text{g})$, $(5.776\pm0.09,\ 3.405\pm0.08,\ \text{and}\ 4.405\pm0.10\ \text{mg/}100\text{g})$ and $(91.97\pm0.13\%,\ 91.95\pm0.11\%)$ and $94.11\pm0.13\%$) respectively.

The total solids values were higher in yogurt fortified with mango juice (16.23±0.02%), pomegranate juice (13.92±0.02%) and plum juice (13.69±0.03%) more than control yogurt (13.53±0.02%). Also, probiotic yogurt fortified with mango juice recorded higher T.S. than probiotic yogurt fortified with pomegranate and plum juices compared to probiotic yogurt. Fat and protein content was lower in probiotic yogurt fortified with plum, mango and pomegranate juices than probiotic yogurt. In general, all the chemical composition parameters of, T.S, fat, protein and ash % are gradually increasing with time of storage.

Yogurt fortified with plum, mango and pomegranate juices had lower pH and higher TTA values when compared with control yogurt. While, probiotic yogurt fortified with pomegranate juice was the lowest in pH and the highest in

acidity more than probiotic yogurt fortified with plum and mango. The obtained data showed that the pH values declined during storage, and the opposite for the TTA values.

2-Rheological Properties

Syneresis

Syneresis refers to the whey that separates from yogurt during the storage period. The results revealed that there are no significant differences in syneresis volume between control yogurt (6.6±0.7 ml) and probiotic yogurt (6.6±0.7 ml). Also, the same manner was found between yogurt fortified with plum juice and probiotic yogurt fortified with plum juice. The addition of 10% plum juice to probiotic yogurt caused the highest syneresis amount (16.2±0.09 ml) compared to pomegranate (7.30±0.6 ml) and mango (6.90±0.7 ml) juices. The syneresis amount increased gradually during the storage period. The probiotic yogurt treatments showed lower syneresis amounts when compared to, mango and pomegranate yogurt respectively

Viscosity

The results indicated that there are no significant differences between probiotic yogurt (165±15 cP), control yogurt (163±11 cP) and probiotic yogurt fortified with mango (161±10 cP) in viscosity. Probiotic yogurt fortified with plum and pomegranate juice showed the lowest viscosity.

3- Sensory evaluation

The data showed that the addition of 10% plum juice to control yogurt decreased body and texture (27±0.26), appearance (13±0.21), flavor (42±0.22) and overall acceptability (91±0.88). Also, the addition 10 % mango juice to control yogurt showed no significant difference between treatments in body and texture and appearance, while fortification of yogurt with mango juice increased the flavor (44.5±0.23) and acceptability (97.5±0.83) and showed to be the most favorable yogurt however, the addition 10% pomegranate juice into probiotic yogurt showed significant decrease in acceptability more than plum juice. All the sensory properties indicators were gradually decreased with storage period up to 14 days.

4- Chemical analysis

Determination of total phenolic, flavonoids contents and antioxidant activity of yogurt fortified with plum, mango and pomegranate juices samples

Probiotic yogurt, showed higher values of antioxidant activity (50.31±0.27%), total phenolic (11.952±1.85 mg/100g) and flavonoid contents (1.952±0.16 mg/100g)

than control yogurt ($46.44\pm0.92\%$, 11.464 ± 1.18 mg/100g and 1.607 ± 0.13 mg/100g), respectively.

Furthermore, probiotic yogurt fortified with 10% plum juice, showed higher values for these parameters than yogurt fortified with plum juice. The same manner was observed for probiotic yogurt fortified with pomegranate juice more than probiotic yogurt fortified with mango juice. Probiotic yogurt fortified with mango juice has the lowest antioxidant activity, total phenol and total flavonoids compared to probiotic yogurt fortified with plum and pomegranate juices. Total phenolic, flavonoid contents and antioxidant activity values are gradually decreased along with storage for all treatments.

Identification of phenolic compounds in juices and yogurt by HPLC

Yogurt fortified with different fruit juices has an increase in phenolic compounds detected by HPLC compared with control yogurt. Gallic acid and chlorogenic acid were the most abundant phenolic compounds in control yogurt, yogurt fortified with mango, probiotic yogurt fortified with mango juice and probiotic yogurt. However, Gallic acid and Catechin were found to be the highly contributed compounds in pomegranate juice, yogurt

fortified with pomegranate, probiotic yogurt fortified with pomegranate and probiotic yogurt fortified with plum juice

However, naringenin was found to be the lowest phenolic compound detected in probiotic yogurt, control yogurt and yogurt fortified with 10 percent plum juice treatments $(0.40\pm0.02,~0.13\pm0.01~\text{and}~0.10\pm0.02~\text{µg/ml})$, respectively. Ferulic acid was found to be the least abundant compound in yogurt fortified with 10 percent mango juice and probiotic yogurt fortified with 10 percent plum juice treatments $(0.16\pm0.01~\text{and}~0.03\pm0.02~\text{µg/ml})$, respectively. Moreover, quercetin was the lowest existent phenolic component in probiotic yogurt fortified with 10 percent mango juice treatment with an amount of $0.10\pm0.01~\text{µg/ml}$.

5- Microbiological analysis

The results indicated that the probiotic bacteria count was the highest count of all total bacterial counts over all treatments. However, the bacterial count of the probiotic yogurt fortified with 10 percent plum juice is the lowest count of all treatments. The addition of mango juice elevated the total viable counts of bacteria in all treatments. However, the addition of pomegranate juice decreased the total viable counts of bacteria in all treatments.