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Original article

Estimation of the Contents of Total Phenols and Antioxidants in Some Tea Samples by Spectrophotometric Method

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Keywords.
Antioxidant, Total Phenols, Tea, Spectrophotometer.

ABSTRACT
Determining how total phenolic and antioxidant levels are distributed among tea samples collected in Al-Bayda, Libya, from different local markets represented the manuscript's principal research aim. Ten distinct samples were bought from various businesses. In this investigation, the spectrophotometric approach was employed. The standard selected for calculating the antioxidant activity was Tannic acid. Measurement of the total phenol content instead relied on the Folin reagent method. The study's findings showed that the amounts of tannins ranged from 13.34 to 59.44 ppm, with the sample of Algafela tea having the greatest content and the sample of Lipton tea having the lowest. The total phenolic contents varied from 24.00 to 59.538 ppm, with the sample of Algafela tea having the greatest amount and the sample of Ahmad tea having the lowest. When compared to black tea samples, green tea samples had the highest total phenolic content and antioxidant capacity.

Introduction
Worldwide consumption patterns place Tea from the Camellia sinensis plant among the leading beverage choices. The six main varieties of this beverage—green, yellow, white, oolong, black, and dark illustrate a range from completely unfermented to post-fermented forms, based on the level of fermentation [1]. Evidence indicates a significant association between habitual Tea intake and a lower risk of chronic diseases, particularly cancer and cardiovascular conditions. While its bioactive components have demonstrated anti-inflammatory, endothelial-enhancing, and antioxidant properties, the precise mechanisms of action remain somewhat unclear [2].
In green tea, antioxidant properties derive mainly from key phytochemicals in the flavanols group, especially catechins (flavan-3-ols) as well as theaflavins [3&4]. In addition to their capacity to neutralize free radicals, these compounds demonstrate significant pharmacological potential, including the regulation of serum glucose, lipid levels, and uric acid concentrations, which are believed to be mediated by the interaction of flavanols with specific cellular transporters and enzymatic pathways. This research study aims to evaluate the number of tannins and total phenolic compounds across all samples selected for the study. A study by the University of Shanghai focused on geographical provenance and observed that 30 samples of green tea infusions from different regions of China displayed significant variability in phenolic content and pronounced antioxidant activity due to geographical position. Tea thus shows great potential to serve as a source of dietary antioxidants of natural origin, while also having potential as a base of functional drinks. Therefore, the teas analyzed in this study are major contributors of phytochemicals to the diet.
While other studies done in Libya have applied analytical techniques such as spectrophotometry, GC-MS and flame photometry to extensively study local plants, there is still a lack of data on commercial tea. Primary emphasis in this study therefore fell on measuring total phenolic content (TPC) together with overall antioxidant capacity across tea samples purchased in local Libyan markets.

Methods
Sampling
Ten samples of black and green tea were gathered from El-Baida City's local markets. Table 1 provides illustrations of the samples.

Sample Preparation
Oven drying was applied to thirty grams of each sample at 105 degrees Celsius, and the treatment continued for two hours. Cooling preceded mortar grinding, after which each sample stayed in polyethylene tubes until required for use.

Solvents Utilized
Petroleum ether and Absolute methanol 96%.

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Table 1. The types of the studied samples

No	Sample
1	ALTAJ tea
2	LYBYANO tea
3	AL ZAHRA tea
4	NABT tea
5	ALgafela tea
6	Lipton tea
7	Al Arose tea
8	Green chine tea
9	Tegan tea
10	Ahmad tea

Solutions Prepared

To prepare Potassium ferricyanide solution (0.008 M), 0.248 grams of solid material were dissolved in 100 ml of distilled water. A 1% FeCl3 Solution was obtained after 1 gram of solid material had dissolved in 100 mL of distilled water.

Preparation of the 0.1 M HCl Solution required transferring 2.08 mL of concentrated solution into one standard 250 mL beaker before adding enough distilled water to complete the designated final volume inside it.

A 20% Sodium Carbonate Solution was made through dissolution of 20 grams of solid material in 100 mL of distilled water.

Standard Tannic Acid Solution

Standard 10 mL beakers received tannic acid solution adjusted to 0, 1, 2, 3, 4, and 5 ppm. After this concentration series had been prepared, absorbance measurements for every level were obtained using 720 nm as the measurement wavelength.

Determination of Antioxidants (as Tannic Acid)

A one-gram powder portion underwent defatting through petroleum ether. During the next stage, two stirring cycles were applied to the defatted powder, each using 10 ml of methanol and 10 ml of a 1% hydrochloric acid: methanol solution, thereby producing successive extracts. After vacuum evaporation of the three combined extracts, 10 ml of methanol was used to dissolve the residue. The addition of three milliliters of diluted water, three milliliters of 0.008 M $K_3Fe(CN)_6$, three milliliters of 0.1 M HCl, and one milliliter of 1% FeCl3 was facilitated by halving the amount of the compound filtrate from the previous steps. The measurement wavelength was 720 nm for absorbance recording.

Determination of Total Phenolic Compounds (TPC)

Extract aliquots were placed in a 10 mL flask before 3 mL of distilled water was added to the flask. A mixture of 0.5 ml of Folin-Ciocalteu reagent (1:1 with water) and 2 ml of 20% sodium carbonate was then introduced. The test solutions were heated for one minute, cooled, and then...

Results

The standard curve of Tannic of standard solutions, which was used to estimate anti-oxidant as Tannic acid was shown in Figure 1. The Lunda max of Tannic acid was obtained at a wavelength of 720nm

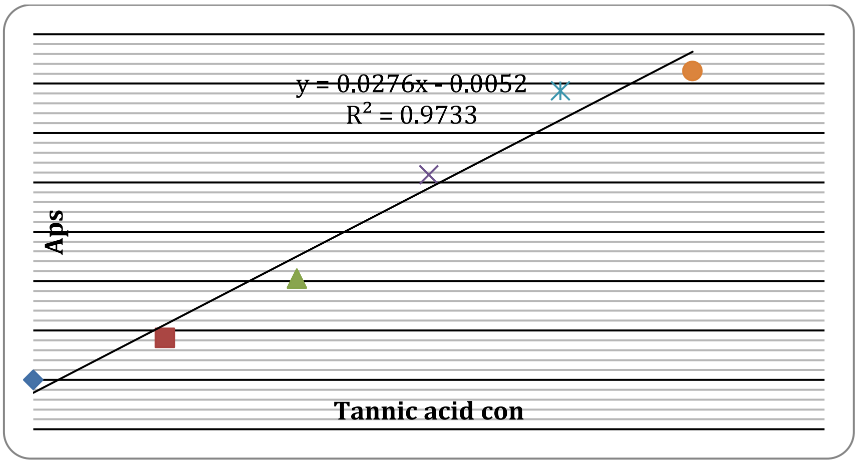

Figure 1. The Standard curve of Tannic acid at (720nm).

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Antioxidant activity in this investigation ranged from 13.34 to 59.44 ppm, with sample S5 (Algafela tea) having the greatest amount and sample S6 (Lipton tea) having the lowest. The findings appear in table (2).

Table 2. The concentration of tannin in tea samples.

Sample No.	Concentration (ppm)
1	28.637
2	20.255
3	32.055
4	58.433
5	59.44
6	13.74
7	34.44
8	42.77
9	38.18
10	30.18

Total phenolic compound (TPC):
The Standard solution measurements at a wavelength of 650nm

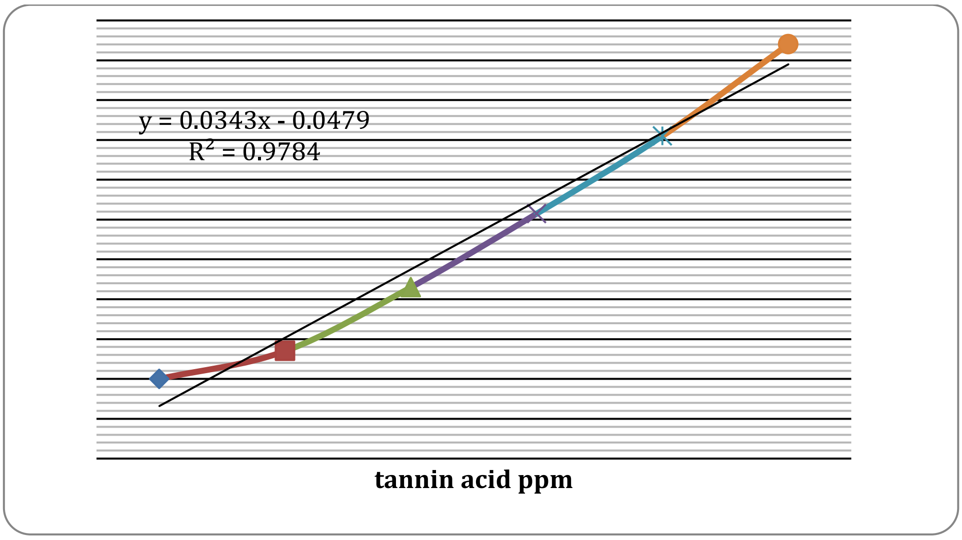

Figure 2. The Standard Curve at λ_{max} (650nm).

Total phenolic contents (TPC) ranged from 24.00 to 59.538 ppm, with sample S5 (Algafela tea) having the greatest content and sample S10 (Ahmad tea) having the lowest (Table 3).

Table 3. Concentration of (Total phenolic Compounds) TPC in tea samples.

Sample No.	Concentration (ppm)
1	32.67
2	31.494
3	41.365
4	33.82
5	59.538
6	34.47
7	29.55
8	45.263
9	31.44
10	24.00

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Discussion

As secondary metabolites with bioactive properties, phenolic compounds naturally occur throughout many plant species; foods such as vegetables and fruits also contain these substances [38]. Their recognized anti-inflammatory and antioxidant effects help explain why consumption of these compounds is linked to many health advantages. Some benefits of reduced susceptibility to chronic and degenerative diseases associated with oxidative stress include decreased risks for cancer, neurological, and cardiovascular disorders. Oxidative stress occurs when there is an imbalance in cellular redox. This imbalance results in an excess of reactive oxygen species (ROS) within the cell and overwhelms the cell's capacitative defense.

Cells produce oxygen metabolites like ROS during essential cellular metabolic processes like respiration and photosynthesis. Excess ROS can adversely alter critical signaling pathways and contribute to the pathogenesis of numerous chronic conditions. On the other hand, ROS at low concentrations is essential for the regulation of cellular metabolic activities. Oxidative stress may occur when ROS is abundant compared to cellular antioxidants. The highly reactive hydroxyl radical, superoxide anion, and hydrogen peroxide may cause covalent modifications to critical cellular macromolecules such as proteins, lipids, and DNA, and such modifications can lead to chronic and degenerative diseases. Phenolic compounds are antioxidants because they can scavenge reactive oxygen species to less reactive or inactive forms and protect the integrity of cells.

Certain phenolic compounds are known to bind to transition metals and activate antioxidant defense systems by regulating the redox state and controlling enzymatic activity. Because of their many therapeutic uses and ability to prevent several diseases, the biological functions and antioxidant potential of different plant phenolics are well studied [41-44]. Numerous environmental studies in Libya have been published on the characterization of water, soil, plant, and marine samples using analytical methods such as AAS, ICP, GC/Mass, and others [45 - 105]. Most of these studies report the chemical constituents present in the measured samples.

Conclusion

The samples contain antioxidant and total phenol contents; the values were higher in green tea compared with black tea. The presence of these compounds is mainly from natural sources; the differences in their contents are mainly due to the black tea processing and the different of tea plant locations.

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