# Spectrophotometric determination of Total Phenol in Apples, Coffee and Tea

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#### **Abstract**

This study aimed to determine the concentration of total phenol in different types of apples (green, yellow and red), tea and coffee from different countries.

Samples were taken from different sources10 samples of each type green, red, yellow, 3 samples of coffee and tea type.

The results shown high concentrations of phenol in all samples of apples, coffee and tea compared with normal value in human body, this increasing of concentration lead to many harmful effect to human health.

#### الخلاصة

تهدف هذه الدراسة إلى تقدير تركيز الفينول الكلي في أنواع مختلفة من التفاح ( الأخضر، الأصفر، الأحمر) وأنواع مختلفة من ( الشاي والقهوة ) من مناشئ ودول مختلفة.
اخذت النماذج من مصادر مختلفة حيث تم اخذ 10 نماذج من كل نوع من التفاح وثلاثة نماذج من كل نوع من الشاي والقهوة.
وجد أن هذه النماذج تحتوي على تراكيز عاليه من الفينول في انواع التفاح وكذلك في انواع الشاي والقهوة مقارنتا مع القيمة الطبيعية للفينول في الجسم مما يؤدي إلى مضار مختلفة لصحة الإنسان.

## Introduction

Phenols are highly toxic and extensively used in various industries, these are readily absorbed by ingestion, inhalation and through intact skin<sup>(1)</sup>.

The main sources of phenolic compounds in the diet are fruits and vegetables, including apples and processed apple products<sup>(2)</sup>. Dietary intake of phenolics is

estimated to be about one gram per day, this is significantly higher than that of all other dietary antioxidants, including vitamin C, vitamin E and carotenoids<sup>(3)</sup>.

Phenolic compounds widely distributed in plants, attract significant scientific interest due to their bio-functional health-promoting properties<sup>(4-7)</sup>. Fruits are potential sources of natural phenolic antioxidants used as food additives for the prevention of lipid oxidation and thus prolongation of food self-life<sup>(8-11)</sup>.

It is not known if phenol causes cancer in humans but cancer developed when phenol was applied to the skin several times per week for the lifetime<sup>(12)</sup>. Apart from being a suspected carcinogen, phenol and some of its derivatives can also be toxic or lethal to aquatic life, when phenol enters the environment it has a half-life in soil between 1 and 10 days, it has a half-life in water between 10 and 30 days, larger or repeated releases of phenol can remain in the air, water and soil for much longer periods<sup>(13,14)</sup>.

Tea and coffee are rich in polyphenols with a variety of biological activities, many of the demonstrated activities are consistent with favorable effects on the risk of chronic diseases, 4-O-methylgallic acid (4OMGA) and isoferulic acid are potential biomarkers of exposure to polyphenols derived from tea and coffee respectively, 4OMGA is derived from Gallic acid in tea, and isoferulic acid is derived from chlorogenic acid in coffee<sup>(15)</sup>.

Tea and coffee are widely consumed beverages, they can be major dietary sources of polyhydroxylated phenolic compounds (polyphenols), including phenolic acids and flavonoids, tea contains primarily flavonoids, as well as up to 15% (25–50mg per cup) total polyphenols present as non-esterified or esterified Gallic acid, coffee polyphenols are almost entirely chlorogenic acid (100–200 mg per cup) (16-18).

This work aim to determine the concentrations of total phenol in different types of apples, coffee and tea.

### **Experimental**

#### **Instrumentals:**

A UV-Probe model (UV-1650) spectrophotometer (Schimadzu-Japan) was used for all absorbance measurements, pH measurements were made with Knick-Digital pH meter (England), Digital Balance, Sartorius, (BP 3015- Germany) and Water bath, Gesellschaft Fur Labortechnik (Germany)

#### **Preparation of Samples:**

Samples were taken from different sources, 10g of apples (10 samples of each type green, red, yellow), 2g of coffee and tea of each sample (3 samples of each type).

Samples were cut to small species and digested with (1:3) perchloric acid to nitric acid mixture<sup>(19)</sup>, and heating by using water bath for 30 min. at a temperature of 50-80 c°, samples were filtered, and measured.

## **Preparation of Reagent**<sup>(20)</sup>:

Carbonate-bicarbonate buffer, 0.05 M, pH 10.1.

Dissolve 3.18 g of anhydrous sodium carbonate plus 1.68 g of sodium bicarbonate in water and dilute to 1 liter.

Solution A: dissolve 0.09 g of 4-aminoantipyrine in 200 mL of carbonate-bicarbonate buffer.

Solution B: dissolve 2.6 g of boric acid and 0.38 g of potassium ferricyanide in water and dilute to 200 ml.

Standard solution:

0.05 g and 0.1 g of phenol per liter of water. Water is used as a blank.

#### **Methods:**

The procedure is as follows:

- (a) To test tubes, add 0.2 mL of sample and 1.5 mL of solution A, and mix well.
- (b) Add 1.5 mL of solution B to the mixture, mix well, and allow standing for 3 min.
- (c) Measure the absorbance of the standard and sample vs. a blank at 500 nm.

The color is relatively stable, but it is preferable to make the reading within 20min.

#### **Results & Discussion**

#### **Calibration Curve of phenol**

A calibration curve of the phenol was obtained by following the proposed procedure that show in experimental part but with out of sample, concentrations of phenol are prepared by taking a known volume from standard solution of phenol and diluted by D.W, ten concentrations was obtained to make the calibration curve, this calibration curve shown in figure (1).

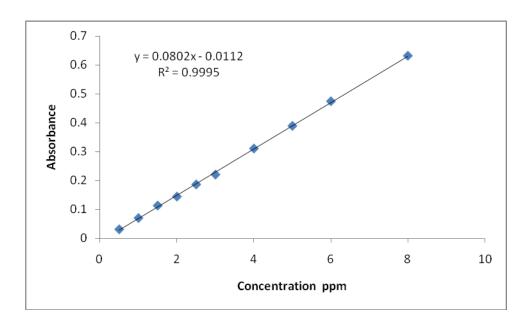


Figure (1) Calibration curve of phenol.

Calibration curve was obeyed in the range (0.5 - 8) ppm, all concentrations were prepared from a standard solution of phenol.

#### Analytical parameters:

Analytical parameters was determined, like detection limit it was (0.0035) ppm, linearity ( $R^2$ ) was (0.9995), correlation factor (r) was (0.9997), molar absorbitivty ( $\epsilon$ ) was 3.22 x 10<sup>4</sup> L.mol<sup>-1</sup>.cm<sup>-1</sup>.

Precision and accuracy of the analytical procedure was determine, it was R.S.D % (2.98) % and  $E_{rel.}\%$  and  $R_e$  % were (-2.62) %, (97.38), these parameters was determine by using the flowing equations

$$S.D = (\sum (Xi-X)^2/(n-1))^{1/2}$$

$$RSD\% = (S.D/X) \times 100$$

$$D.L = (C \times S.D \times 3) / X$$

$$E_{rel} \% = (u-d) / u$$

$$R_e = 100 + E_{rel}$$

Xi is the absorbance

X is the average of absorbance

n is the number of samples

C is the concentration

u is the true value

d is the analytical value

The concentration of the phenol in all samples were determined by using the calibration curve.

### **Apples**

The average concentrations of the phenol (ppm) in different samples of apples were shown in table (1).

Table (1) the average concentrations of phenol (ppm) in apples

	` '	<u> </u>	
Countries	Green Apple	Red Apple	Yellow Apple
USA	4.866	4.936	4.894
Turkish	4.796	4.810	4.781
China	4.743	4.753	4.008
Iran	4.511	4.585	4.557

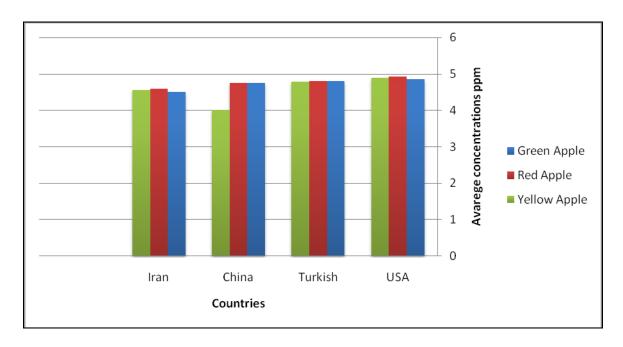


Figure (2) average concentrations of phenol in apples samples

## Black tea

The average concentrations of the phenol (ppm) in different samples of black tea were shown in table (2).

Table (2) the average concentrations of phenol (ppm) in tea

Countries	Con. ppm
India	1.673
Bangladesh	1.462
China	2.025
Iran	2.035
Vietnam	2.967
Kenya	1.561
Pakistan	1.645
Cirelanka	1.715

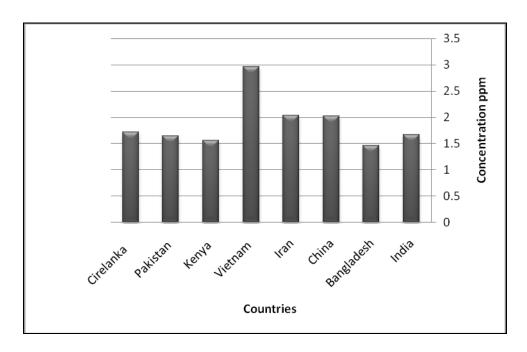


Figure (3) average concentrations of phenol in tea samples

#### Coffee

The average concentrations of the phenol (ppm) in different samples of coffee were shown in table (3).

Table (3) the average concentrations of phenol (ppm) in coffee

Countries	Con. ppm
Egypt	5.800
KSA	1.631
Arabic	2.869
Brazil	4.556
Vietnam	2.967
Turkish	1.322

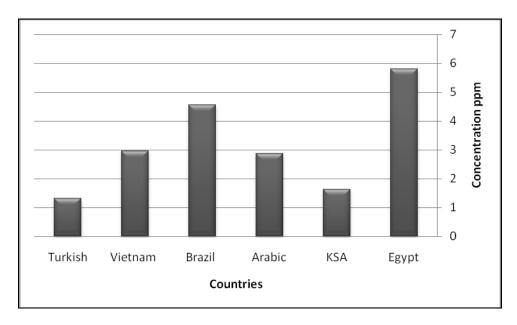


Figure (4) average concentrations of phenol in coffee samples

From tables 1 to 3 the average concentration of total phenol in apples, coffee and tea was very high compared with the acceptable value in human body  $(\approx 0.28 \text{ppm})^{(21)}$ .

Apple, tea and coffee are a plants these plants need many complementary factors to grow up and give a good product, these factors are soil, water, air, ground water and fertilizer, these factors may be contaminated by phenolic compounds from different sources like factories, industries pesticides, dust, bad time storage due to accumulation of contamination on the products. In industries, phenols are important chemicals, widely used in industries for the manufacture of products such as dyes, insecticides, disinfectants, wood preservatives, in other hands as chemical products in building, agriculture and hospital<sup>(22)</sup>. Fertilizer contained chemical components contained many phenolic compounds these compounds can contaminated the product by phenol.

Dusts and bad storage time may be contaminated the product by phenolic compounds from air pollutants.

Moreover the occurrence of phenols in the environment stems from the production and use of numerous pesticides, in particular phenoxyherbicides like 2.4 dichlorophenoxyacetic acid or 4-chloro-2-methylphenoxyacetic acid (MC PA) and also phenolic biocides like pentachlorophenol (PCP), dinoseb or diarylether

Pesticides, the presence of phenols in the ecosystems is also related with production and degradation of numerous pesticides and the generation of industrial and municipal sewages. Some phenols are also formed during natural processes (23-25).

High value of phenol in apples, tea and coffee lead to many hazardous for human body because phenol is well absorbed from the gastrointestinal tract and through the skin of both animals and humans. It is metabolized principally by conjugation (by sulfation and glucuronidation) with a minor oxidation pathway leading to quinone-related reactive intermediates which bind covalently to protein and are detoxified by conjugation with glutathione. Most of the absorbed phenol and its metabolites are excreted in the urine, with trace amounts of excreted in expired air and the feces<sup>(26)</sup>.

In addition, very small amounts of phenol is produced endogenously as a breakdown product of protein metabolism by the action of bacteria on normal constituents of the diet in the gut and excreted independent of external exposure to the compound. Some of this internally-produced phenol may be eliminated in the feces and some may pass to the blood, phenols and their derivatives commonly exist in the environment<sup>(27)</sup>.

Phenol irritates skin and causes its necrosis, it damages kidneys, liver, muscle and eyes. Damage to skin is caused by its coagulation related to reaction to phenol

with amino acids contained in keratin of epidermis and collagen in inner skin<sup>(28)</sup>.

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