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COMPARATIVE ANALYSIS OF HUMIDITY, DRY MATTER, PH AND SOD OF SOME APPLE SORTS

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ABSTRACT

Apples are a great source of vegetable substances (quercetin, catchiness). They are products with a low glycemic index and high antioxidant odds. Aim of the study: to examine some physical and chemical characteristics of the following apple sorts: Aidere, Jonah Gold, Chadeo, Mutsu and Florina. Methods applied: 1. Systematic approach and critical analysis of the available scientific periodicals. 2. Mathematical and statistical methods. 3. Technical Analysis / spectrophotometric method. Results: The dry content in % (percents) ranges from 12,62 ± 0,43 (Chadeo) to 16,70 +_ 0,26 (Jonah Gold); pH (average) - from 3.41 (Mutsu) to 4.23 (Florina); Antioxidant activity in % - from 65,77 (Aidere) to 42,63 (Florina).

Keywords: antioxidant activity, dietary properties, low glycemic index

INTRODUCTION

In mythology, the apple is a well-known symbol of love, marriage, youth, fertility, longevity and immortality. The apple is one of the best fruit for the human organism, due to its diverse chemical composition. Besides being a nutritious food, this fruit processes considerable and valuable medicinal and dietary properties. The apple fruit is considered to be one of the richest in its varieties. It is believed that there are over 10000 apple varieties in the world but in practice, varieties differ by: taste of the fruit; coloring of the fruit skin; time for full ripeness. Based on their taste, the apple varieties are divided into two large groups: varieties with sweet fruit constitution; varieties with sour fruit constitution.

According to the coloring of the fruit skin the varieties of apples are: yellow and red. All of the varieties that have yellow or green-colored skin at camp maturity correspond to yellow; The reds include all those with red-colored fruit skin, regardless of whether the red color covers the whole fruit or is only on spots, stripes or flaming.

Apple refers to the family Rosacea, subfamily Pomoida, kin Malus Mill.

We have over 7,500 officially registered cultivated varieties of apples and the number of all types - is hardly immeasurable. Each year the world grows more than 5.5 million tons of apples. Globally, the largest producers are: China, USA, Iran, Turkey, Russia, Italy, and India. With a huge variety of species and varieties, apples are widespread in almost all parts of the world; each variety has different characteristics relating to their size, color, taste, flowering period and characteristics of the fruit.

Apples are a great source of vegetable substances (quercetin, catchiness). They are products with a low glycemic index and high antioxidant odds [1,2,3,4,5].

The aim of the research was: to examine the pH value, the antioxidant activity and the amount of the dry content of the following apple sorts: Aidere, Jonah Gold, Chadeo, Mutsu and Florina.

- methods and apparatus for determination; Results analysis.

EXPERIMENTAL

Time of the study: one year study that spans from October 2015 till October 2016

Place of study: Faculty of Pharmacy, Medical University Plovdiv, Bulgaria, College of Medicine, Plovdiv, Bulgaria and Trakia University, Stara Zagora, Bulgaria

Objects of the survey are the following sorts of apples: Aidere, Jonah Gold, Chadeo, Mutsu, Florina bred in the villages Novo selo and Byaga situated in the Upper Thracian Valley in Bulgaria.

Based on a sensory analysis (color and appearance), the apples are in a fully mature stage, with no physical integrity violations.

Scientific methods applied:

- Systematic approach and critical analysis of the available scientific periodicals.
- Mathematical and statistical methods

- Technical Analysis / spectrophotometric method

Apparatus:

UV-VIS spectrophotometer „Thermo Scientific“

Electromagnetic stirrer „Vortex 2 Genie “

Centrifuge „Centronix Microcentrifugo 1236V“

Juicer „Philips“

Drying cabinet: LABORBIO LABSTER 16020

Analytical Scale: Denver Tools

Ph meters: WTW INOLAB PH-720 and DENVER INSTRUMENT

ULTRA BASIS PH

Chemical reagents used:

Hipoxantin oxidase

Phosphate buffer

RESULTS AND DISCUSSION

Of the apple sorts considered, we have defined the following characteristics:

Determination of moisture and dry content (AOAC, 945.32).

Samples of fresh apples are cut into pieces and then homogenized with a blender to a homogenous mass.

Working procedure:

1. The empty weighing panes are incinerated in a desiccator at 105° C for 2 hours, and then transferred in a desiccator to cool down. Weigh the empty weighing panes with the covers (M2) on an analytical balance.

2. Weigh, on an analytical balance, about 10g of the fresh sample in each weight glass (M1). Separate the sample evenly throughout the bottom for a more uniform drying.

3. Samples are dried for 4 hours at 105 ºC.

4. After drying, the weighing glasses, altogether with the partially closed lids, are transferred to the desiccator to cool down. Reweigh the weighing glasses together with the dried samples (M3).

5. Calculate the percent moisture, respectively, and the percentage of dry matter of formula (1) and formula (2).

$$Moisture\ (\%) \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (1)$$

$$Dry\ matter\ (\%) = 100 - Moisture\ (\%) \ldots\ldots (2)$$

Where:

M1 – The mass of the fresh sample and the weighing glass, g

M2 – The mass of an empty weighing glass, g

M3 – The mass of the dried sample and the weighing glass, g

Table 1: Determination of moisture and dry matter of different varieties of apples

Number	sort of Apple	Dry content (%)	Moisture (%)
1	Chadeo	12,62 ± 0,43	87,38 ± 0,43
2	Florina	14,62 ± 0,50	85,38 ± 0,50
3	Mutsu	13,79 ± 0,23	86,21 ± 0,23
4	Jonah Gold	16,70 ± 0,26	83,30 ± 0,26
5	Aidere	14,55 ± 0,20	85,45 ± 0,20

Defining pH with two types of pH meters: WTW INOLAB PH-720 and DENVER INSTRUMENT

Table 2: Ultra basis pH

	pH c WTW INOLAB pH-720	pH c DENVER INSTRUMENT ULTRA BASIS PH	pH (mean value)
Mutsu	3,42	3,40	3,41
Florina	4,22	4,25	4,235
Chadeo	4,00	4,10	4,05
Jonah gold	3,85	3,75	3,80
Aidere	3,51	3,49	3,50

One of the natural solutions to the problem with the free radicals are antioxidants [5,6,7]. Apples combine flavonoids, fiber and antioxidants in a unique and incomparable way.

The experiment is based on the ability of antioxidants (products with antioxidant properties) to trap free radicals - super oxide anion (super oxide radical). The mechanism of formation of the super oxide radical is the following:

$$Hypoxanthine + hypoxantine\ oxidase + uric\ acid + O_2 super\ oxide\ radical \quad \textbf{(3)}$$

$$O_2^0 + O_2^0 + H_2 \rightarrow \xrightarrow{SOD-superoxide\ dismutase} \rightarrow H_2O_2 + O_2 \quad \textbf{(4)}$$

We evaluate the degree of antioxidant activity by the difference in the degree of staining of the solution.

The degree of staining is determined by UV-VIS spectrophotometer.

We determine the antioxidant activity by the difference of absorption of the control sample and the sample with the corresponding apple juice.

To determine the antioxidant activity of each apple variety we use 3 tubes.

"0" - zero tube

"K" - a control tube

"1" - test tube

We obtain nectar from apple by squeezing them prior the experiment using a PHILIPS juicer.

To prepare the juice for the experiment we centrifuge it for 5 minutes at 14000 rpm in centrifuge Centronix Microcentrifugo 1236V. The goal is to settle the nectar particles and to remain pure juice.

In all tubes we put in 1,5ml kit (hypoxanthine + nitrobluetetrasole) [6,7,8,9].

In tube, "0" we set 100µl sample (juice) + 400 µl buffer solution (phosphate buffer) instead of hypoxanthine.

In test tube "K" we put 400µl hypoxanthine oxidase +100 µl buffer instead of sample.

In test tube "1" we put 400µl hypoxanthine oxidase +100 µl sample.

Tube "0" is used to remove the difference in color.

Tube "K" has the highest activity of free radicals.

Tube "1" has a lower activity of free radicals, as some of them are caught by the sample.

Sequence of actions:

Mix the three tubes with electromagnetic stirrer VORTEX GENIE 2 for 10 seconds.

Put the tubes in a water bath for 10-15 minutes at 380C.

Put the tubes in cold water for 5 seconds to stop the reaction.

Determination of the activity of the samples with a UV-VIS spectrophotometer/ Thermo Scientific. Working length λ = 560 nm [10,11,12,13].

First we measure the control tube with maximum activity.

For our 4 cases (3 varieties of apples) we measure 3 controls and take the average. The controls are the same [14].

$$1{,}5ml\ kit + 400ml\ hypoxanthine\ oxidase + 100ml\ buffer \quad \textbf{(5)}$$

Super oxide dismutase:

1 – 1.528 nm

2 – 1.450 nm

3 – 1.379 nm

Average value: 1.4523 nm

The difference in the three measurements should be no greater than 30%

We measure the absorbance of the nine samples "1". For each we make two measurements and take the average (Table 2).

Table 3: Absorption of test tube 1

Apple sorts / village	Measurement 1	Measurement 2	A tube.1	ΔA = A мax.- A tube.1
Mutsu village Byaga	0.714	0.611	0.662	0.790
Florina village Byaga	0.823	0.844	0.833	0.619
Chadeo village Novo Selo	0.782	0.763	0.772	0.680
Jonah Gold village Novo Selo	0.952	0.835	0.893	0.559
Aidere village Novo Selo	0.489	0.504	0.497	0.955

Before each measurement, we reset the apparatus with a solution of test tube "0", to avoid loss of the absorbance due to the coloration [13,14].

For antioxidant activity we judge by the change in absorbance ΔA between tube "K" and tube "1".

Antioxidant activity (or SOD like activity) is calculated by the following formula:

$$SOD = \frac{\Delta A}{\frac{A_{max}}{2}} . dilution = \frac{\Delta A.2}{A_{max}} . \frac{2}{0,1} = \frac{\Delta A.2.2}{A_{max}.0,1} = \frac{\Delta A}{A_{max}.40} \quad \textbf{(6)}$$

For 0,1 ml juice:

$$SOD = \frac{\Delta A.2}{A_{max}} . \frac{2}{0,1} \quad \textbf{(7)}$$

2ml is the full volume of the tube, and 0,1 – the volume of the juice sample.

In each tube „K" and „1" we have the following volume:

$$1,5ml\ kit + 0,1ml\ tube + 0,4ml\ SOD = 2ml \quad \textbf{(8)}$$

$A_{max} \rightarrow\ A\ of\ the\ tube\ "K"$

$\Delta A - A_{max} \rightarrow A\ of\ the\ tube\ "1"$

In % :

$Amax = 100\%$

And

$tube\ 1 = X\%$

where X are the percents of absorption of the tube.

The subtraction 100 % - X % results in antioxidant activity measured in %, which is given respectively for each variety in Table 3.

Table 4: Antioxidant activity of the apple sorts

Apple sorts / village	SOD	ANTIOXIDANT ACTIVITY IN %
Mutsu village Byaga	**21.76**	**45.59**
Florina village Byaga	**17.05**	**42.63**
Chadeo village Novo Selo	**18.73**	**46.83**
Jonah Gold village Novo Selo	**15.40**	**61.50**
Aidere village Novo Selo	**26.31**	**65.77**

CONCLUSIONS

The careful reading of the available scientific periodicals and the conducted data in our experiments allows us to make the following conclusions:

1. There is no dependence between the dry and moisture content and the SOD /i.e. the antioxidant activity/**.**

2. UP to a certain extent there is a direct relationship between the pH and the SOD / i.e. the antioxidase activity/.
3. Apples are a products with high antioxidant activity

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