# STUDY ON THE MODIFICATION OF THE OXIDATIVE STATUS OF AN *ECHINACEA AUGUSTIFOLIA* TEA WHEN USING SWEETENERS

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**Abstract**

*Echinacea tea reduces the risk of contamination with respiratory viruses.*

*Echinacea tea comes from the daisy family and offers a sweet and potent floral flavour. This herbal tea may help boost immune health and fend off the common cold. Echinacea helps maintain healthy skin regeneration and reduces the burning sensation in cases of injury.*

*This working paper presents one valuable study regarding the influences that occur during the sweetening task.*

*In this case, maintaining the best balance between the reduced and oxidized forms of enzymatic cofactors - for the specific oxidoreductases - is one of the important aim.*

*The moving of the gravitational centre of redox balance to the oxidized area can be the source for transforming the valuable bio-compounds in toxic unwanted compounds. For this reason, the monitoring of the redox balance in this functional food is mandatory.*

*This paper presents a personal method to determine the best natural or synthetic sweetener that can be used without any problems when sweetening the Echinacea tea from a free contamination area.*

***Key words**: **Echinacea Tea, NAD, FMN, sweeteners -***

## INTRODUCTION

There are three main types of Echinacea plants used in the tea industry: *Echinacea purpurea, Echinacea pallida and Echinacea angustifolia*. Echinacea angustifolia has large leaves and purple flowers, while the pallida variety has flowers that are more of a pale pink colour. The *Echinacea Purpurea* variety has reddish-purple flowers and is known in Europe as an important immune system stimulant [3]

The substances that are extracted from the root of the plant are different from those that are extracted from the upper parts of the plant. Thus, the roots contain more volatile oils, flavonoids, amides and glycoproteins, compounds that contribute to strengthening the immune system, while the aerial parts contain more polysaccharides [1].

In addition to these important nutritional compounds, Echinacea also contains:

proteins; fatty acids, fat-soluble vitamins (A and E) and water-soluble B vitamins; iron, copper, potassium, calcium, resins, polysaccharides [1].

In addition to these important nutritional compounds, Echinacea also contains:

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Studies show that Echinacea has numerous benefits for the consumer's

body, and Echinacea tea can be considered an important functional food. Thus, the tea can be used successfully in strengthening the immune system, preventing cancer, as an analgesic, in relieving respiratory problems, in preventing microbiological infections and improving oral health, in combating digestive problems, in regulating oxidative status, in preventing cardiovascular diseases [3]..

If the Echinacea plant is harvested from contaminated areas, or through processing it creates the conditions for toxicity which can lead to allergic reactions, digestive disorders, nausea, severe fatigue, even anaphylaxis. Failure to maintain a balance in processing between the concentrations of oxidised and reduced forms of some coenzymes of oxidoreductases can lead to serious nutritional diseases and diabetes.

In this case, it is very important to monitor the concentrations of FMN- and NAD-dependent enzymes, the ratio of oxidised and reduced forms of which causes important changes in the redox potential value of the tea and affects the health of the consumer [5-8].

Flavonoids have been found to have antidepressant activity. Exist more of studies which evaluate the antidepressant activity of flavonoids in Echinacea flowers with possible involvement of monoamines [3].

Also, the antioxidant (bioflavonoid) content of Echinacea leaves has anti-inflammatory activity and helps reduce fat deposits installed on the arteries, prevents cell degradation, being also helpful in people with type II diabetes [4].

The regular tea consumption also helps the maintain bladder health and prevents constipation, supports the health of the digestive system in general [10-11].

The use of Echinacea flowers from a Protected Natural Area, from non-polluting areas or from areas where is practiced the organic farming is greatly for consumers, the risk of heavy metals being accumulated in the edible part being near zero [6]. Such systems are capable to produce the high-quality agricultural raw materials – which can be used to obtain the highly demanded dietary supplements [12-14].

For growers of these plants, knowing of eco-pedological practices is essential in obtaining the high value added foods that can be used both in current nutrition and in the prevention or control of certain diseases [6].

Also, the use of certain techniques for lyophilizing the edible parts of Echinacea plants can preserve - in high concentrations and for long periods of time - the properties of antioxidants in these plants [11-12].

## MATERIALS AND METHODS

In this experiment, for to obtain the tea from *Echinacea purpurea* flowers was used one of pure plant infusion by adding 10-15 flowers to one liter of boiling water. After 20 minutes (the established period), one of unsweetened version of *Echinacea* tea (control variant) could be obtained.

In order to know the level of purity of the plants, the lack of pollution indicators by contamination with heavy metals and / or minerals in excess, it was used one atomic absorption spectrometry method after the plant samples had been corresponding mineralized.

For this experiment it was used only the herbal tea made from unpolluted and uncontaminated *Echinacea*. This tea can be used like as blank to constitute the control experimental variant. This zero initial pollution / contamination it is necessary to be able to prevent rapid oxidation of certain processes or change of antioxidants concentration - to obtain the best results in manage process of sweetening tea.

In the first step of sample preparation for atomic absorption spectrometry (AAS) it was used the mineralization process (microwave digestion). Microwave digestion is used to prepare samples of all types (plant) for elemental analysis by AAS, which require the sample to be in the form of a solution in

order to introduce it into the analyser. Acid digestion is employed to break down the sample matrix leaving the elements of interest in solution and ready for analysis. For this purpose it used a CEM Mars system of microwave mineralized, 1200W. The MARS system of CEM is a multimode platform equipped with a magnetic stirring plate and a rotor that allows the parallel processing of several vessels per batch. Method: Briefly, was weighed with analytical precision 10 g dry substance (d.s.). For the mineralization was added to each digestion cartridge 10.00 g product (Echinacea tea), 5 mL of concentrated nitric acid and 3 mL of 30% hydrogen peroxide. For blank was used one digestion cartridge without product, just reagents.

For AAS method was used one Varian SpectrAA 220Z Atomic Absorption Spectrometer Furnace System with Varian SpectrAA 220Z Auto Sampler , Varian GTA 110Z Furnace, Varian UltrAA and afferent Windows interface software.

In order to study the oxidizing level of agricultural products, was determined the enzymatic activity and the ratio between the oxidized forms and the ones reduced for FMN (Flavin Mono nucleotide) and NAD (Nicotinamide adenine Dinucleotide). [9].

In this paperwork there are shown the values of the ratios between oxidized and reduced forms for both FMN - dependent enzymes and also NAD – dependent enzymes for the experimental variants of Echinacea tea. The concentrations of oxidized forms and reduced forms of FMN and NAD from experimental variants of Echinacea tea were obtained using molecular absorption spectrophotometer, specific calibration curves for these enzymatic cofactors – for this kind of vegetal and the necessary temperature corrections. To verify the maximum of molecular absorption spectra in the UV - Vis range for NAD, NADP (Nicotinamide Adenine Dinucleotide Phosphate), FMN, were used the Pure Analysis chemicals and Single Addition Methods.

For samples spectroscopy UV-VIS used a spectrophotometer UV-VIS T92+, product by PG Instruments; with high performance double beam spectrophotometer available with a fixed (2nm) or variable (0.5, 1, 2, 5nm) spectral bandwidth, spectral range of 190-1100nm, Wavelength accuracy +/- 0.3nm. During the lab tests the specific repeatability, accuracy and robustness conditions were accomplish, respecting all requirements according to all present standards.

After the control variant was formed, the following experimental variants were made by adding sweeteners:

$V_1$- unsweetened Echinacea tea (RS = reference sample);

$V_2$- Echinacea tea with sugar sweetener;

$V_3$- Echinacea tea with saccharine sweetener;

$V_4$- Echinacea tea with Aspartame sweetener (from Equal product);

$V_5$- Echinacea tea with Cyclamate sweetener;

$V_6$- Echinacea tea with Zuckli sweetener;

$V_7$ - Echinacea tea with Sucrazit sweetener;

$V_8$ - Echinacea tea with honey from Acacia.

The saccharine had been produced by SICOMED (19 mg saccharine/tablet)- and had been added into boiled tea.

The Aspartame was by NUTRASWEET (Equal Brand Sweetener) (90 mg Equal/tablet), Cyclamate (100 mg Natrium Cyclamate/tablet) was from S.C.ARMEDICA S.A., Zuckli (40 mg Cyclamate + 4mg Saccharine/ tablet ), had been produced by BERLINER CHEMIE, Sucrazit (Natrium bicarbonate 59.52%, Saccharine 23.81%, fumaric acid 16.67% for one tablet) had been produced by BISCOL CO LTD (from Elite). The natural acacia honey was added in Echinacea tea under $50^{o}$ C (dropped below 50 degrees Celsius for temperature of tea).

## RESULTS AND DISCUSSIONS

The obtained results from atomic absorption spectrometry (both for dry matter in the plant and for the 1:10 aqueous extract) are shown in Table 1.

The table shows that there were no improved concentrations of heavy metals or ion concentrations that could act on Echinacea antioxidants. As can be seen from the results of the reports of the concentrations of the oxidized and reduced forms of the experimental variants both inside the tea (Figure 1) and in the liquid interface (Echinacea tea) / air, the smallest changes - induced by the sweetening process - are recorded at $V_8$ (experimental variant using honey) and $V_5$ (experimental variant with cyclamate).

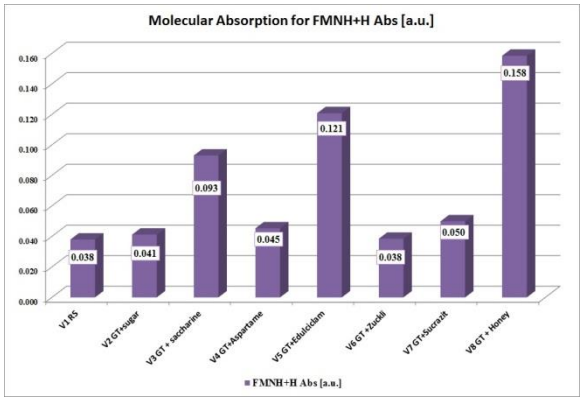



Figure 1- Value of FMNH+H + absorption. for experimental variants

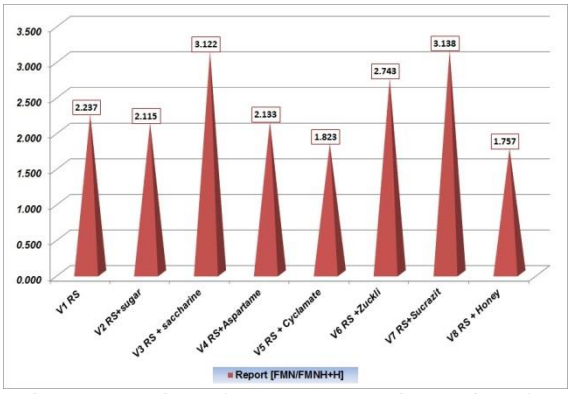



Figure 2- Value of NADH+H + absorption for experimental variants

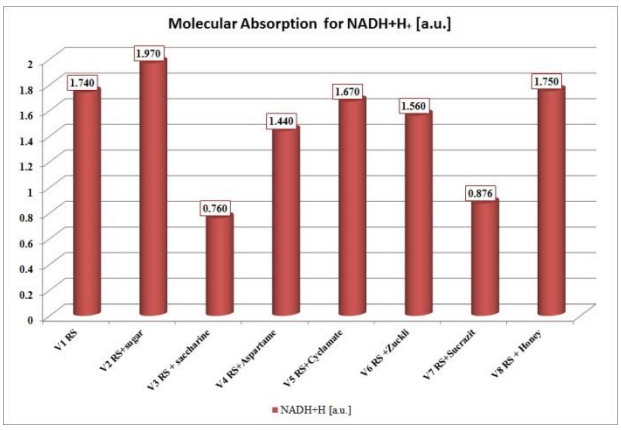
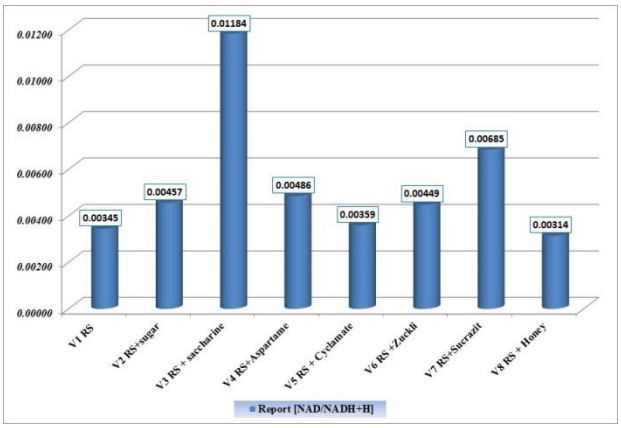



Fig.3 - Value of Report FMN/FMNH+H + for experimental variants




.Fig.4 - Value of Report NAD/NADH+H + for experimental variants

Table 1- Atomic Absorption Spectroscopy for Echinacea Tea Base Line

| Indicators | Dry Matter ppm (mg/kg) | Aqueous extract 1:10(mg/L) | Comment |
|---|---|---|---|
| $Na^{+}$ | 9.125 | 0.463 | - |
| $K^{+}$ | 27.250 | 2.319 | - |
| $Ca^{2+}$ | 77.500 | 1.263 | - |
| $Mg^{2+}$ | 45.750 | 3.250 | - |
| $Zn^{2+}$ | 17.125 | 0.338 | - |
| $Mn^{2+}$ | 10.750 | 0.525 | - |
| $Fe^{2+}$ | 14.250 | 0.213 | - |
| $Al^{3+}$ | 0.075 | 0.001 | - |
| $Cu^{2+}$ | 0.769 | 0.009 | - |
| $Pb^{2+}$ | 0.026 | 0.001 | - |

## CONCLUSIONS

After analysing the obtained results, the following conclusions were drawn:

- To design and build the best experimental variants of Echinacea tea, starting from a blank (untreated and unpolluted) version of Echinacea it is necessary to control the heavy metals and certain minerals in the base plant. Without this mineralization of the samples and verification by atomic absorption spectrometry, the results obtained from the sweetening can be unsatisfactory,
- Taking into account the reactivity of heavy metals, the strong activity of some metal ions, the aggressiveness of some free radicals, the increase of this reactivity during technological processes of additive (in this case, of sweetening) it is necessary for the plants - which used in the design of Echinacea teas like functional foods - to be free of contamination and not to passed through additional processes that allow the breaking of certain valuable chemical compounds in unwanted radicals,
-- Using reagent-free techniques - such as Atomic Spectroscopy and UV-Vis Spectroscopy, combined with electrochemical analysis methods (also verified by monitoring the concentrations of the main redox agents that act on the inside of liquid and/or the tea - air interface) it is possible to obtain very valuable results - which can be real indicators in setting up the best experimental variants of nutrition and treatment for certain classes of consumers,
-- The best experimental variant that is used by healthy consumers for preventive purposes is an experimentally sweetened variant with acacia honey like as sweetener ($V_8$),
- The best experimental variant – which can addressed to consumers with nutrition, hyperacidity and / or diabetes problem is an experimentally sweetened variant with of sodium cyclamate ($V_5$)
-- The experimental variant of Echinacea tea-sweetened with saccharine produced the most significant changes in active and reduced-form concentrations of FMN and NAD – relative to the similar ones of the unsweetened variant and can be considered as a variant to be avoided.


## ACKNOWLEDGEMENTS

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