RESEARCH ON THE DEVELOPMENT OF FUNCTIONAL FOODS BASED ON BLACK TEA

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Abstract

Consumers are increasingly demanding functional foods on the market due to growing awareness of the close link between diet and health. The development of functional foods has made considerable progress in recent years, and interest in this type of food is growing because, in addition to their nutritional value, they also have beneficial effects on human health.

Black tea is one of the most widely consumed beverages in the world. Its chemical composition is notable for its high content of polyphenolic compounds, such as theaflavins and thearubigins, as well as amino acids, alkaloids (especially caffeine), and essential minerals. The main redox processes that take place are primarily the oxidation of catechins—phenolic compounds present in fresh leaves—which lead to the formation of thearubigins and theaflavins. Experiment has demonstrated a clear correlation between the activity of some natural and synthetic sweeteners and the change in the concentrations of oxidized and reduced forms of some redox agents. The aim of the research is to identify the changes produced in the specific redox processes of these supplements. The best experimental variant was the one that produced the fewest redox imbalances. Two optimal variants were selected for both consumers without health problems and consumers with health problems, such as diabetics.

***Key words**: functional foods, sweeteners, black tea, redox processes*

INTRODUCTION

The development of functional foods has made considerable progress in recent years, and interest in this type of food is growing because, in addition to their nutritional value, they also have beneficial effects on human health. These types of foods have the capacity to reduce the risk of certain diseases and maintain the body's physiological functions at an optimal level. Consumers are increasingly demanding functional foods on the market due to growing awareness of the close link between diet and health.

In nature, we can find sources of valuable chemical compounds, but black tea is a particularly important natural source of bioactive compounds, making it suitable for use in functional foods.

Black tea is one of the most widely consumed beverages in the world. Its chemical composition is notable for its high content of polyphenolic compounds, such as theaflavins and thearubigins, as well as amino acids, alkaloids (especially caffeine), and essential minerals. These chemical compounds have well-known biological activity, antioxidant, antimicrobial, and anti-inflammatory properties. The potential of black tea to become a basis for the development of functional foods is due to its versatility and the fact that it can be easily accepted from a sensory perspective. The process of making black tea involves complex enzymatic fermentation, during which several redox (oxidation-reduction) reactions take place that transform the natural polyphenols in *Camellia sinensis* leaves into new compounds with different structures and functions.

The main redox processes that take place are primarily the oxidation of catechins—phenolic compounds present in fresh leaves—which lead to the formation of thearubigins and theaflavins. Oxidative enzymes, such as polyphenol oxidase (PPO) and peroxidase (POD), play a central role in the conversion of natural polyphenols (especially catechins) into theaflavins and thearubigins. These chemical compounds give black tea its characteristic color, specific taste, and powerful antioxidant activity.

Among the redox enzymes involved are those dependent on redox cofactors such as NAD^+ (nicotinamide adenine dinucleotide) and FMN (flavin mononucleotide). Nicotinamide Adenine Dinucleotide (NAD) is a widespread coenzyme in all living cells. One such nucleotide contains an Adenine base and a Nicotinamide base. NAD is present in living cells in two forms: one oxidized and one reduced, abbreviated as NAD+ and NADH+H+ (H stands for Hydrogen). Flavin Mononucleotide (FMN) or Riboflavin-5 Phosphate is a bio-molecule produced from Riboflavin (vitamin B2) by the enzyme Riboflavin-Kinase and functions as a prosthetic group for several types of oxidoreductases. These coenzymes play an essential role in electron transfer reactions, ensuring intracellular redox balance and participating in the regeneration of oxidized or reduced compounds during fermentation. NAD- and FMN-dependent enzymes, such as dehydrogenases and flavoprotein oxidoreductases, indirectly assist in the oxidation of polyphenols, either by generating hydrogen peroxide that is subsequently used by peroxidases or by regulating the redox potential of tea leaf cells.

In order to maintain the bioactive properties of the final product, it is important to understand the role of NAD–FMN oxidoreductases in the biochemical processes that occur during the fermentation of black tea for the optimization of technological processes.

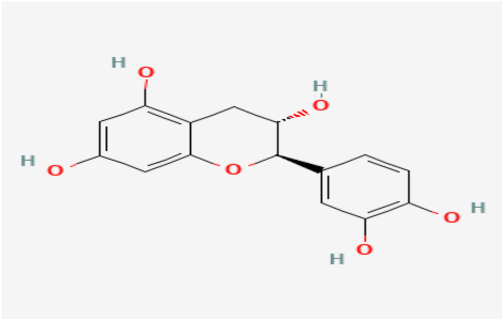

Figure 1. The molecular structure of catechin

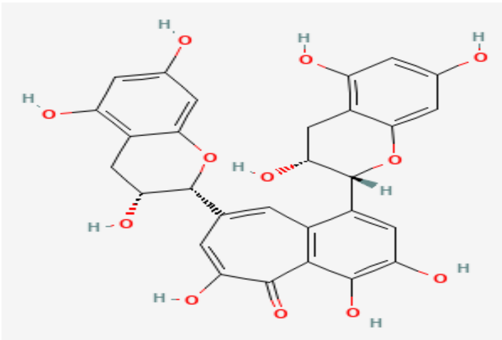

Figure 2. The molecular structure of theaflavin

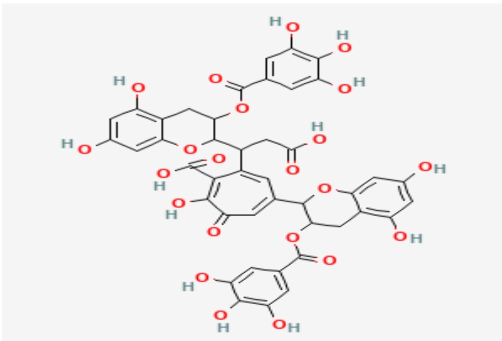

Figure 3. The molecular structure of thearubigin

MATERIALS AND METHODS

The effect of various natural and synthetic sweeteners on the chemical composition of black tea was determined through laboratory analyses.

In order to obtain the control variant (V_0) of unsweetened black tea, 10 grams of black tea leaves were added to one litre of water in a suitable container. The tea was then subjected to a heating, cooling, and filtration process to produce the experimental variant from which the other sweetened black tea variants were derived.

Figure 4. Experimental variants

A total of ten varieties of sweetened tea were obtained from this experimental variant (control variant). The following description presents the varieties of black tea with additives:

V_0: control sample of unsweetened black tea;

V_1: black tea sample sweetened with white sugar;

V_2: black tea sample sweetened with brown sugar;

V_3: black tea sample sweetened with honey;

V_4: black tea sample sweetened with saccharin;

V_5: black tea sample sweetened with Sucrazit;

V_6: black tea sample sweetened with Diamond;

V_7: black tea sample sweetened with fructose;

V_8: black tea sample sweetened with xylitol;

V_9: black tea sample sweetened with sorbitol;

V_{10}: black tea sample sweetened with stevia.

The experimental variant Sucrazit (V_5) is the trade name of a synthetic sweetener containing 25% saccharin, citric acid, whose acidity has been buffered with sodium bicarbonate.

The experimental variant (V_6) Diamond is the trade name of a synthetic sweetener containing a combination of sodium cyclamate and sodium saccharin.

As shown in our previous studies, a 2% dilution was used to allow the incident light beam to pass through the black tea solution. This dilution was obtained by passing the black tea through filter paper and then transferring it to the spectrophotometer cuvettes for analysis.

To obtain the molecular absorption spectra of the experimental variants, a T92 Plus UV-VIS spectrophotometer from PG Instruments (UK) was used. The spectrophotometer was configured to operate at a wavelength bandwidth of 1 cm and to record molecular absorption values from nanometer to nanometer in both the visible range (400-700 nm) and the UV range (190-400 nm).

The equipment automatically records spectral curves, switching between deuterium and tungsten lamps at 361 nm through automatic programming. To verify the values obtained, the T92 Plus spectrophotometer was set to perform an automatic re-evaluation for each measurement.

Special parallelepiped UV quartz cuvettes with a square side of 1 cm were used to measure absorption.

Prior to analysis, the raw material was tested using A.A.S. (atomic absorption spectroscopy) to check for contaminants such as residues or heavy metals. The effect of sweeteners on the basic chemical composition was investigated using UV-VIS optical spectrometry and mathematical statistics.

The analysis was facilitated by Windows-based software. Results are expressed as mean ± standard error of the mean (SEM), except for high-oil plants, which are expressed as mean ± standard deviation (SD). For a better statistical analysis, two-way Kruskal-Wallis ANOVA followed by Dunn's post-hoc test can be used.

All calculations were performed using ORIGIN PRO 2020 software.

RESULTS AND DISCUSSIONS

When developing innovative functional foods based on black tea, it is important to ensure that the sweeteners used preserve the active forms of the main chemical compounds.

The results of this stage of research showed the best sweetening option for black tea-based functional foods. The product that was manufactured and chosen for use (as a raw material) in the next stages of the process had a chemical composition similar to that of the unsweetened product, but also much better sensory properties.

The use of the AAS technique revealed high concentrations of potassium, magnesium, and calcium in the parts of the tea plant used, as shown in Table 1.

Table 1. Cations composition of black tea obtained by AAS technique

Indicator/ Constituent	**BLACK TEA**	
	Dry matter ppm (mg/kg)*	**Watery extract 1:10 ppm (mg/L) Average value**
Na^{+}	1012±3.56	153.04
K^{+}	16720.2±3.26	285.21
Ca^{2+}	742±2.46	7.98
Mg^{2+}	462±1.64	7.93
Zn^{2+}	48±2.48	1.79
Mn^{2+}	84.5±1.89	0.23
Fe^{2+}	74.24±1.84	0.68
Al^{3+}	10.42±0.67	0.49
Cu^{2+}	10.46±0.66	0.35
Pb^{2+}	0.036±0.001	missing

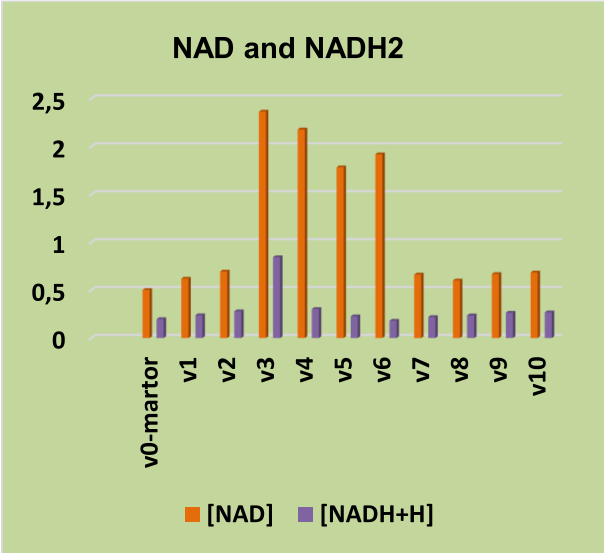

Figure 5. The NAD and NADH2 concentration

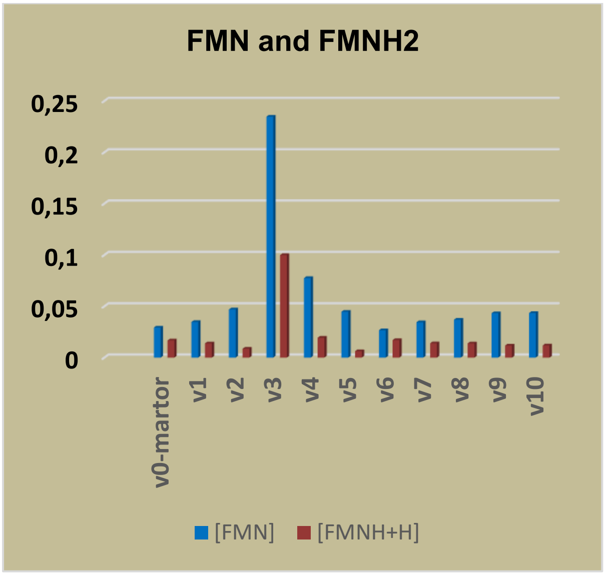

Figure 6. The FMN and FMNH2 concentration

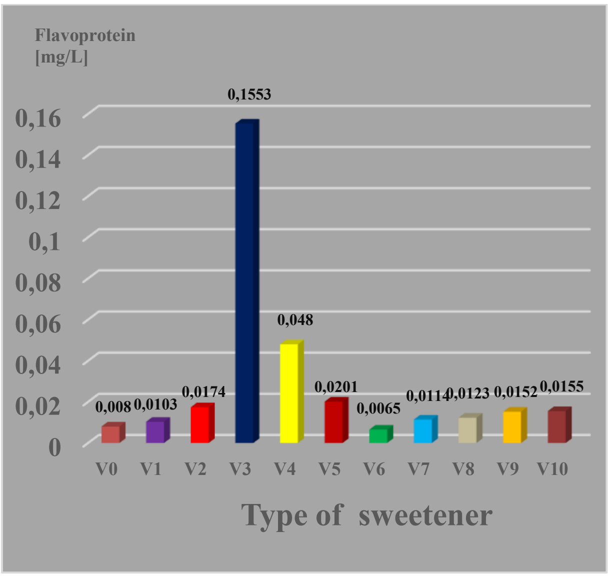

Figure 7. Black tea flavoprotein concentration

In the experimental variant V_0, the control variant, a peak in NAD+ was observed, accompanied by a decrease in tocopherol and a high content of theobromine.

When white sugar is added to the experimental variant V_1, minor changes in pigments and a decrease in certain chemicals, especially theobromine, can be observed. The tendency for flavoproteins to change is also observed when brown sugar is used in V_2, but to a greater extent in terms of protecting oxidized and reduced forms.

The concentrations of oxidized and reduced forms are higher in brown sugar than in white sugar.

Honey used as a sweetener in the V_3 variant showed the highest concentrations of oxidized and reduced forms, as shown in the adjacent graphs.

The version of the experiment in which saccharin V_4, was used as a sweetener protects the oxidized and reduced forms.

Sucralose (V_5) has a significant influence on the concentration of primary active compounds present in black tea.

The use of Diamond V_6 results in the lowest concentrations of flavoprotein forms, as can be seen from the graph.

Studies have shown that the use of a mixture of sodium cyclamate and saccharin can help maintain the concentration of certain oxidoreductase coenzymes stable. This also prevents them from losing their powerful antioxidant properties.

In the case of variant V_7, fructose was used. It should be noted that this has no significant effect on color intensity in the UV and visible spectrum, thus ensuring that the color tone is maintained. The levels of oxidized and reduced forms are kept low.

The use of sweeteners such as xylitol (V_8), sorbitol (V_9), and stevia sweetener (V_{10}) is mandatory to keep the levels of oxidized and reduced forms at a constant level.

Sorbitol and stevioside have similar effects on flavoprotein concentrations. There is a similar situation with regard to the influence of sweeteners on the concentrations of reduced forms.

CONCLUSIONS

It is evident that a comprehensive analysis of the results obtained and an in-depth interpretation of the values provided will facilitate the direct derivation of several significant conclusions.

It has been demonstrated that adding honey to black tea as a sweetener induces a higher oxidation state, which leads to a change in the clarity of the tea.

Sucralose has been shown to facilitate consumer use by acting as a buffer in the active core of saccharins, thus inducing a sweet taste under conditions of reduced oxidability. The use of sodium cyclamate and saccharin mixture ensures a certain degree of preservability of the concentration of the hydrogenated forms of specific oxidoreductase coenzymes while preserving their strong antioxidant character (V_6). The addition of brown sugar, despite its effect of altering color intensity and hue, serves to protect reduced forms. Honey and saccharin variants have been shown to be responsible for the highest levels of turbidity, which may have a visual impact on customers.

White sugar (V_1) and xylitol (V_8) are the best sweetener choices because they are the closest to the control variant.

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