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Research Article

Whole wheat flour enriched with nectarine powder - antioxidant activity, microbiological and moisture sorption characteristics

Adelina Vasileva¹✉, Albena Durakova², Hristo Kalaydzhev³, Milena Dimitrova-Dicheva⁴, Zhivka Goranova⁵, Kliment Georgiev¹

¹ Department of Mechanical and Instrument Engineering, Faculty of Mechanical Engineering, Technical University of Sofia, Branch Plovdiv, 25 Tsanko Diustabanov Str., Plovdiv 4000, Bulgaria

² Department of Process Engineering, Technical Faculty, University of Food Technologies – Plovdiv, 26 Maritsa Blvd, 4002 Plovdiv, Bulgaria

³ Department of Analytical Chemistry and Physical Chemistry, Technological Faculty, University of Food Technologies – Plovdiv, 26 Maritsa Blvd, 4002 Plovdiv, Bulgaria

⁴ Department of Milk and Dairy Products Technology, Technological Faculty, University of Food Technologies, 26 Maritsa Blvd, Plovdiv, 4002, Bulgaria

⁵ Institute of Food Preservation and Quality - Plovdiv, 154 Vasil Aprilov Blvd., 4003 Plovdiv, Bulgaria

Abstract

The aims of the current scientific study are to present the initial data research on the flour mixture of whole wheat flour enriched with nectarine powder of Bulgarian origin – intended for sponge cake production - was created. The natural content of the antioxidants was evaluated through the *in vitro* methods DPPH, ABTS, FRAP, and CUPRAC. The equilibrium moisture content (EMC) and monolayer moisture content (MMC) were investigated at 10°C, 25°C, and 40°C, resembling the diapason of temperature conditions in the storage house. The water activity (from 0.1 to 0.9) is provided through saturated salt solutions of LiCl, CH₃COOK, MgCl₂, K₂CO₃, MgNO₃, NaBr, NaCl, and KCl. The obtained adsorption and desorption EMC declare the tendency – with its increase, the temperature decrease at the constant value of *a_w*. According to Brunauer's classification, sorption isotherms are of the second class, showing their characteristic S-shape profile. The modeling of the obtained sorption data includes their mathematical description using one of the three-parametrical models of modified Halsey, Oswin, Chung-Pfost, and Henderson. The Brunauer-Emmett-Teller linearization was presented to the values of the monolayer moisture content – for the adsorption process they are between 5.78% and 8.94%; and for the desorption - between 7.25% and 8.89%.

Keywords

nectarine, whole wheat, flour, mixture, antioxidant, sorption isotherms, monolayer moisture content

Abbreviations DPPH – 1,1-diphenyl-2-picrylhydrazyl radical; ABTS – 2,2'-azinobis (3)-ethylbenzthiazoline-6-sulfonic acid; FRAP – Ferric reducing antioxidant power; CUPRAC – Cupric reducing antioxidant capacity; EMC – equilibrium moisture content; MMC – monolayer moisture content; BET – Braunauer-Emmett-Teller

✉ Corresponding author: Adelina Vasileva, Chief Assist. Prof. in Department of Mechanical and Instrument Engineering, Faculty of Mechanical Engineering, Technical University of Sofia, Branch Plovdiv, 25 Tsanko Diustabanov Str., Plovdiv 4000, Bulgaria, tel: +359 897449031 E-mail: adelina.bogoeva@abv.bg

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Introduction

Nowadays, society is increasingly focused on finding natural sources of bioactive compounds. Obtaining them helps to reduce oxidative stress with the body as well as helps in the fight against some chronic degenerative diseases. A natural enhancement of the immune response against seasonal, influenza, and viral conditions that disrupt a person's normal daily life is sought (Burton-Freeman et al. 2017; Dhalaria et al. 2020; Maheshwari et al. 2022).

Nectarine is a seasonal fruit and ripens in the months from May to September. In the past, it was called "*Persian plum*" and "*naked peach*" because of its characteristic smooth and shiny skin. We associate nectarines with juiciness, a bouquet of aromas, and an explosion of flavours. Apart from being fresh, we can find it canned in the form of compotes, purees, or jams. Its aroma is an integral part of the main flavours, aromas, and essences in products such as ice cream, biscuits, and beverages. In addition to its strong sensory and organoleptic susceptibility, the nectarine has several benefits when consumed fresh. It has a low glycaemic index, and its carbohydrates slowly raise blood sugar, making it suitable for consumption by diabetics. The summer fruit has a low caloric content (from 44 to 65 kcal.100g⁻¹ depending on the variety), contains fiber, vitamins A, B, C, D and E, as well as minerals potassium, phosphorus, copper, magnesium, calcium and zinc. Nectarine has curative and prophylactic effects due to its rich nutritional composition and is recommended for use in cardiovascular problems, improving venous circulation and blood vessel wall tone. Due to its high fiber content, it helps to improve the functioning of the gastrointestinal tract (Tomás-Barberán et al. 2013; Bento et al. 2020; Guo et al. 2020; Mihaylova et al. 2021; Tekgül 2021).

In turn, wheat obtained by grinding whole grains (husk, endosperm, and germ) - wholemeal flour, also has a rich set of micronutrients such as iron, copper, zinc, selenium, magnesium, and vitamins B and E. Despite its low consumer susceptibility, this type of wheat flour is rich in fiber, which promotes faster satiety when consumed and is often recommended when preparing diets (Bressiani et al. 2017; Gómez et al. 2020). The combination of different flour products with increased nutritional values meets consumer requirements and demands.

The development of new combinations of enriched flour mixtures with regional food products would contribute to the development, both scientifically and in business (Mancebo et al. 2016; Bogoeva 2020; Zlateva et al. 2022).

The moisture content of flour mixtures plays an important role in determining the optimal storage conditions to maintain the quality characteristics of the product. The study of adsorption and desorption processes provides this important information (Zhang et al. 2017; Labuza and Altunakar 2020; Fontana and Carter 2020).

Following a literature review, no data was found on the creation of a flour mixture of whole wheat flour with nectarine flour of Bulgarian origin. This gave us the reason to conduct the current study, which determines the physico-chemical characteristics, antioxidant activity, microbiological contamination, and sorption characteristics of a new flour mixture intended to produce sponge cakes.

Materials and Methods

Materials. A flour mixture of 70% whole wheat flour (with an initial moisture content of 11.4%) and 30% dried and powdered nectarines (with an initial moisture content of 14.19%) was composed. Nectarine fruits and whole wheat flour (made by Goodmills Bulgaria LTD) were purchased in local various stores in Plovdiv, Bulgaria. Fresh selected nectarine fruits were sliced until the thickness of 3.0mm ± 5.0mm. Before being finely ground with a blender into powder, the nectarines were dried in a heat pump dryer for 8h at 42°C or until the moisture content reached the range of 13.5% to 14.5%. It was obtained approximately 114.8g±11.48g dried product from 1 kg of fresh nectarines. Preparation was made at the Institute of Food Preservation and Quality - Plovdiv, Bulgaria.

Methods. Chemical analysis (approximate composition) and antioxidant activity of flour mixture. The selected standard methods were used for chemical characterization of samples: Ash content was determined by ICC Standard No. 104/1 (International Association for Cereals Science and Technology 1990). The Kjeldahl method was used to determine total nitrogen content in the samples and the results are multiplied by 6.25 to convert them to crude protein (AOAC 1990). Total lipids and crude fiber were evaluated by standardized

methods (ISO 11085:2015; ISO 5489:1981). Carbohydrates were determined by AOAC Method 988.12 (44.1.30). Moisture content, % - standard method by drying 5 g of flour at 105°C to a constant weight, according to AOAC 960.39, 1990 (AOAC 1990; Palamthodi et al. 2021). All reagents used in the study were of analytical grade.

Antioxidant activity was determined through four different methods – DPPH, ABTS, FRAP and CUPRAC, described in detail by Ivanov et al. (2014), and by Bogoeva et al. (2017).

Microbiological analysis. Total count of mesophilic aerobic and facultative anaerobic microorganisms, according to BDS EN ISO 4833-1:2013; Yeasts and molds, according to BDS EN ISO 21527-2:2011; *Escherichia coli* according to BDS EN ISO 16649-2:2014; *Salmonella* spp., according to BDS EN ISO 6579-1:2017; Coagulase-positive staphylococci, according to BDS EN ISO 6888-1:2022; Coliforms, according to ISO 4832:2006.

Sorption characteristics and stat data. A gravimetric-static method was used for the determination of adsorption and desorption equilibrium isotherms, according to Project COST 90 (Wolf 1985) and Bell and Labuza (2000). The determination of equilibrium moisture content and experimental scheme are described in detail by Bogoeva (2020). The analysis of obtained sorption data was investigated through modified three-parametric models of Oswin, Chung-Pfost, Henderson and Halsey, according to the used formulas as in the research of Bogoeva (2020). The linearization of the Brunauer-Emmett-Teller equation was performed for the calculation of monolayer moisture content (Brunauer et al. 1938). A detailed method for determination, description and estimation of sorption characteristics is reported by Bogoeva (2020).

Results and Discussion

Biochemical characteristics (approximate composition). The physicochemical parameters of the studied mixture of whole wheat flour enriched with nectarine powder were characterized as follows: protein – 10.30±0.48%; total lipids 1.84±0.12%; total carbohydrates 70.50±1.65%; ash 3.20±0.59% and crude fibers 9.46±0.10%. Results are calculated on a dry matter basis: 91.82±0.22%.

Antioxidant activity was verified by four methods differing in mechanism of action: DPPH, ABTS, FRAP, and CUPRAC. Extraction of a 1 g sample yielded 26.48% extract. Tabular data is presented in mMTE.g⁻¹ extract and mMTE.g⁻¹ dw.

Based on the studies conducted, the antioxidant activity of the flour mixture was confirmed by all four selected methods. According to Eskicioglu et al. (2016) cereals such as wheat, rye and oat naturally contain antioxidant dietary fibers.

Table 1. Antioxidant activity of whole wheat flour enriched with nectarine powder through DPPH, ABTS, FRAP and CUPRAC methods

Method	mMTE.g ⁻¹ extract	mMTE.g ⁻¹ dw
DPPH	9.05±0.07	2.40±0.03
ABTS	1.95±0.01	0.52±0.01
FRAP	0.90±0.03	0.24±0.01
CUPRAC	2.83±0.09	0.75±0.02

The nectarine powder in this study was prepared from whole fruit (flesh and skin). According to Gil et al. (2002), the fruit skin contains a higher amount of antioxidants, which complement the antioxidant activity of whole wheat flour. Moreover, nectarines are a good source of pyridoxine, niacin, thiamine and pantothenic acid as well as some electrolytes (Mitic et al. 2016). Overall, the mixture of nectarine powder and whole wheat flour has increased nutritional value and may exhibit different properties in dough formation and quality of baking products (Hu et al. 2021).

Microbiological analysis. Flour is a low-water-activity product that is considered microbiologically safe. Although harmful bacteria are not sustained under such low water activity, foodborne bacteria and fungi can survive for long periods of time (Forghani et al. 2018). The total count of mesophilic aerobic and facultative anaerobic microorganisms, yeasts and molds, coliforms, coagulase-positive staphylococci, *Escherichia coli*, and *Salmonella* spp., was carried out to assess the microbiological quality of whole wheat flour enriched with nectarine powder. The results are presented in Table 2. The total count of mesophilic aerobic and facultative anaerobic microorganisms indicates the general

microbiological quality and hygienic status of any food sample (Khanom et al. 2016).

Table 2. Microbiological characteristic of whole wheat flour enriched with nectarine powder

Microbiological parameters	Results
Total count of mesophilic aerobic and facultative anaerobic microorganisms, cfu.g ⁻¹	7.7×10 ⁵
Molds and yeasts, cfu.g ⁻¹	1.6×10 ⁴
Coliforms, cfu.g ⁻¹	3.0×10 ⁵
Coagulase-positive staphylococci, cfu.g ⁻¹	<10
<i>Escherichia coli</i> , cfu.g ⁻¹	<10
<i>Salmonella</i> , in 25 g	Absent

In the present study, their count was found to be 7.7×10⁵ cfu.g⁻¹. A slightly high count of coliforms, 3.0×10⁵ cfu.g⁻¹, and yeasts and molds, 1.6×10⁴ cfu.g⁻¹, was observed. This may be due to environmental contamination, poor processing or unfavorable storage conditions.

It is well known that flour is a raw material that is brought to consumer readiness by the technology of baking and/or cooking, and this reliably destroys pathogenic bacteria for humans. Pathogenic flora such as *Salmonella* spp., coagulase-positive staphylococci and *E. coli* were not detected in whole wheat flour enriched with nectarine powder.

Sorption characteristics.

The moisture content of the flour mixture was 8.94% d.b. This initial value is reduced to 7.39% d.b. by P₂O₅ for the adsorption experimental analysis and hydrated up to 21.79% d.b. for desorption process for 10 days.

The equilibrium moisture content (EMC) of the adsorption and desorption processes is presented in the form of graphical isotherms in Fig. 1 and Fig. 2. According to the graphical analysis, the obtained isotherms have their own indicative S-form that is typical for the second class of Brunauer’s classification. The EMC values demonstrate the known trend – as sorption capacity rises, the temperature decreases in the condition of constant water activity. That is valid for both sorption processes (Brunauer et al. 1938; Troller and Christian 1978; Iglesias 2012; Kapsalis 2017; Labuza and Altunakar 2020; Bogoeva 2020; Durakova 2020).

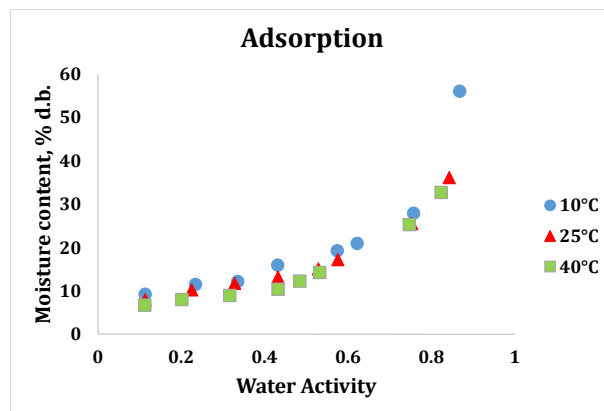


Figure 1. Sorption isotherms at 10°C, 25°C and 40°C for adsorption process

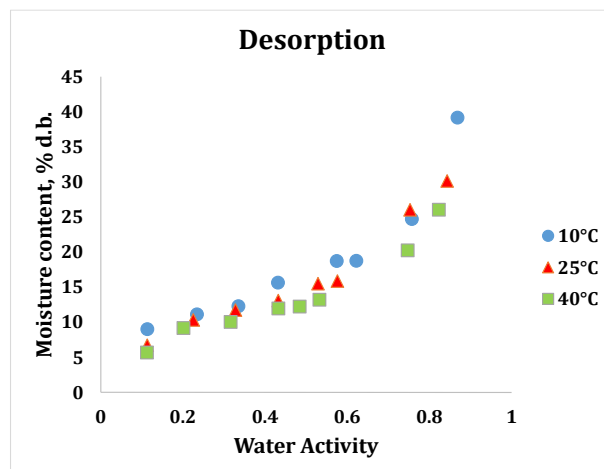


Figure 2. Sorption isotherms at 10°C, 25°C and 40°C for desorption process

The coefficients A, B and C (obtained from the modified three-parametrical Oswin, Halsey, Henderson and Chung-Pfost models), average relative error (P, %), moisture standard error (SEM), and correlation coefficient (R) for adsorption and desorption process are presented in Table 3 and in Table 4 as follow.

The coefficient values A, B and C are evaluated using the *Statistica* 12.0 software by a non-linear regression algorithm. A comparison between calculated and predicted equilibrium moisture content was performed through criteria of model evaluation and suitability – average relative error (P, %) and moisture standard error (SEM) (Muzaffar and Kumar 2016; Labuza and Altunakar 2020; Bogoeva, 2020; Durakova, 2020; Durakova et al. 2022; Statistica, 2012). According to the selected

models, the modified Halsey show the lowest P and SEM values for both sorption processes, and consequently, we recommend this three-parametric model for the most adequate model describing the sorption isotherms.

The monolayer moisture content (MMC) must be investigated in order to predict the optimal storage conditions (temperature and relative humidity) and provide information about the most stable state of the product. Using the obtained sorption data

(within the range of water activity under 0.5) and linearizing the BET model (Brunauer et al. 1940; Bogoeva 2020; Durakova 2020; Durakova et al. 2022) it was calculated the MMC, corresponded to the most stable state of the product. The BET monolayer moisture content (% d.b.) of flour mixture at 10°C, 25°C and 40°C for adsorption process are - 8.94%, 7.65% and 5.78%. Furthermore, the MMC for desorption process are 8.89%, 7.92% and 7.25% at 10°C, 25°C and 40°C.

Table 3. Adsorption model coefficients (A, B, C), average relative error (P, %), moisture standard error (SEM), and correlation coefficient (R)

Model	A	B	C	P	SEM	R
Oswin	19.071284	-0.144812	0.552258	12.93	2.62	0.976
Halsey	4.5044917	-0.016198	1.649599	5.97	2.77	0.992
Henderson	0.0001607	2.6990796	1.866363	18.62	6.17	0.899
Chung-Pfost	302.27168	0.1067663	54.27992	14.04	5.73	0.969

Table 4. Desorption model coefficients (A, B, C), average relative error (P, %), moisture standard error (SEM), and correlation coefficient (R)

Model	A	B	C	P	SEM	R
Oswin	18.063690	-0.116494	0.417070	6.25	1.25	0.989
Halsey	5.0128306	-0.013777	1.876093	5.19	0.98	0.991
Henderson	0.0001530	2.594146	1.914213	17.14	3.92	0.896
Chung-Pfost	380.07870	0.125730	55.40470	8.19	2.59	0.982

As shown from results, the trend is also valid here, the same such as the equilibrium moisture content – with an increase in the temperature, the monolayer moisture content decrease (Bell and Labuza 2000; Muzaffar and Kumar 2016; Labuza and Altunakar 2020; Bogoeva, 2020; Durakova, 2020; Durakova et al. 2022). Summarizing the information obtained on the sorption characteristics, we can recommend keeping the moisture content within the range of 5.78% to 8.94% for optimal diapason providing the longest shelf life and the stable state of product.

Conclusions

It was created and investigated the flour mixture of whole wheat flour enriched with nectarine powder – the source of bioactive compounds. Antioxidant capacity was confirmed using the methods DPPH, ABTS, FRAP and CUPRAC. The microbiological quality and safety were proven by the absence of *Escherichia coli*, *Salmonella* spp., and coagulase-

positive staphylococci). The other microbiological parameters, such as the total count of mesophilic aerobic and facultative anaerobic microorganisms, coliforms, yeasts and molds are under permissive norms. The sorption characteristics, including the equilibrium moisture content and monolayer moisture content, were determined to recommend the optimal conditions of storage for the new flour mixture assuring the most stable state of the product. According to these results, we confirmed the trends that the sorption capacity increase with a decrease in temperature at constant water activity condition, valid for EMC and MMC. The modified three-parametric model of Halsey was the most suitable model for the description of the obtained S-shaped isotherms, based on the lowest values of criteria average relative error (P, %) and moisture standard error (SEM). The MMC values were calculated through the linearization of the Brunauer-Emmett-Teller model and the obtained results for

the adsorption process are: at 10°C – 8.94%, at 25°C – 7.65%, and at 40°C – 5.78%, and for desorption: at 10°C – 8.89%, at 25°C – 7.92%, and at 40°C – 7.25%.

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