## Oat Flour Enriched with Nectarine Powder of Bulgarian Origin – Antioxidant Activity, Microbiological and Moisture Sorption Characteristics

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### Abstract

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Two regional food products were evaluated to create a flour mixture intended for the production of sponge cake. The functional mixture flour consists of oats flour and nectarine powder of Bulgarian origin. The current scientific research presents the physicochemical composition, antioxidant activity, microbiological, and sorption characteristics of the new flour mixture. The performed analysis on the physicochemical parameters were protein 9.98 g, total carbohydrates 74.40 g, total lipids 3.71 g, ash content 3.49 g and crude fibers 6.21 g, calculated on a dry matter basis of 90.82%. The antioxidant activity was determined using the DPPH, ABTS, FRAP, and CUPRAC methods. All microbiological parameters, namely the total count of mesophilic aerobic and facultative anaerobic microorganisms, yeasts and molds , *Escherichia coli*, *Salmonella* spp., coagulase-positive staphylococci, and coliforms, were under permissive norms and no pathogenic organisms were detected. Sorption characteristics, including equilibrium moisture content and monolayer moisture content, were investigated at three different temperatures – 10°C, 25°C and 40°C and water activities in the range of 0.1 to 0.9 following the static-gravimetric method. Estimation of obtained sorption isotherms was performed with a modified three-parametric model of Halsey, Oswin, Henderson and Chung-Pfost, evaluating with criteria of suitability models, namely average relative error (P, %) and moisture standard error (SEM). BET equation was linearized for obtaining the monolayer moisture content values.

Keywords: oats; nectarine; flour mixture; antioxidant activity; sorption isotherms; monolayer moisture content

#### Introduction

Profounding the research of local food products would enrich the national database. That helps to find and rediscover products with improved nutritional facts (qualities and properties) that help fight some degenerative diseases and/or inhibit the development of others (Shin & Hancer, 2016; World Health Organization, 2019). One of them is the nectarine. The typical and highly recognizable smell, color, shape, and taste of fresh fruit attract the appetites of young and old. Nectarine is a variety of peach, often called "naked peach" due to the lack of moss, but with a smooth and shiny peel, resembling that of plums. Young children are often caught with purees made from it due to the many useful rich nutrients such as high fiber, vitamins A, B, C, D, and E, iron, phosphorus, calcium, potassium, copper, zinc, manganese, etc., including the presence of bioactive compounds. As you grow older, it is recommended to take more products rich in antioxidants. Research proves the benefits of consuming nectarines by adults on the cardiovascular system, anemia, hypertension, atherosclerosis, and diabetics due to their low caloric content (between 44 kcal and 65 kcal per 100 g product) and the slow rise in blood sugar, which in turn contributes to the therapeutic and prophylactic action of the product (Tomás-Barberán et al., 2013; Redondo et al., 2017; Xi et al., 2017; Tiwari et al., 2022).

Oats belong to the family of cereals (*Poaceae*) and are a major cereal crop material, which is characterized by a rich nutritional composition and a good ratio of their main macronutrients – carbohydrates, fats, proteins, and ashes. The potential of oats and their derivatives – flakes, flour, and semolina is related to their therapeutic and prophylactic action when consumed. Oats are known as a strong immunostimulant with high antioxidant activity. They have a good effect on people suffering from cardiovascular and kidney diseases, diabetes, infections, and help with the digestive system (Rasane et al., 2015; Paudel et al.,2021; Bouchard et al., 2022).

Technological processing such as drying, storage, and transportation are important stages in the production of flour products. In order to obtain preliminary information on the compilation of the different types of regimes, it is necessary to determine the sorption characteristics of the tested product. Hygroscopic products are always in direct and/or indirect contact with the surrounding temperature and relative air humidity. Therefore, the determination of equilibrium and monolayer moisture content can prevent possible microbiological development and show in what range of moisture content the product should be stored so as to preserve its quality characteristics for as long as possible (Iglesias et al., 2022; Panyathitipong et al., 2022; Zhang et al., 2022).

Based on the literature research, nowadays no data has been found on the study of sorption equilibrium moisture isotherms and monolayer moisture content of a flour mixture of oat flour and nectarine powder.

#### **Materials and Methods**

#### **Raw Material**

A flour mixture of 70% oat flour and 30% dried and powdered nectarines was composed. The study used oat flour produced by Ecosem Bulgaria LTD and purchased in local food market in Plovdiv, Bulgaria. The physicochemical composition of oat flour for 100 g had already been determined by the manufacturer: energy value of 1690 kJ/404 kcal; 9.1 g fat, of which 1.6 g saturated; 70.0g carbohydrates, of which 0.8 g sugars and 14.7 g protein. Nectarine fruits are purchased in local fresh stores in Plovdiv, Bulgaria. The fresh selected nectarine fruits are sliced until a thickness of 3.0±0.5 mm and dried in a heat pump dryer for 8h at 42°C and finely grounded with a Nutribullet blender at the Institute of Food Preservation and Quality - Plovdiv, Bulgaria. The physicochemical composition of nectarine powder per 100 g was determined to be: total lipids 0.25±0.04 g; total carbohydrates 86.33±0.31g; crude fibers 2.13±0.08 g; protein 1.98±0.16 g and ash content 4.87±0.02 g, calculated on a dry matter basis: 94.13±0.78% for nectarine flour.

# Physicochemical parameters, antioxidant activity and microbiological load

Well established standard methods were used for the chemical characterization of samples. Ash content is determined by ICC Standard No. 104/1 (International Association for Cereals Science and Technology, 1990). The Kjeldahl method is 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 are evaluated by standardized methods (ISO 11085, 2015; ISO 5489, 1981). Carbohydrates are 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 (AOAC, 1990). Determination of antioxidant activity using the methods DPPH, ABTS, FRAP, and CUPRAC was performed as described by Ivanov et al. (2014) and by Bogoeva et al. (2017). Determination of the total number of mesophilic aerobic and facultative anaerobic microorganisms according to BDS EN ISO 4833-1: 2013; Determination of yeasts and molds, according to BDS EN ISO 21527-2: 2011; Determination of Escherichia coli according to BDS EN ISO 16649-2: 2014; Determination of Salmonella spp., according to BDS EN ISO 6579-1: 2017; Determination of coagulase-positive staphylococci according to BDS EN ISO 6888-1:2022; Coliforms - ISO 4832:2006.

#### Sorption Characteristics and Stat data

In order to identify the sorption characteristics, the static gravimetric method was applied, as has also been done by Bejar et al. (2012), Hidar et al. (2018), Iglesias et al. (2022), Panyathitipong et al. (2022), Bogoeva & Durakova (2020) and Bogoeva (2020). The most common temperature ranges at the storage house are  $-10^{\circ}$ C, 25°C and 40°C. The saturated eight salt solutions of LiCl, CH<sub>3</sub>COOK, MgCl<sub>2</sub>, K<sub>2</sub>CO<sub>3</sub>, MgNO<sub>3</sub>, NaBr, NaCl and KCl aid in obtaining a water activity range (a<sub>w</sub>) from 0.1 to 0.9. To describe the sorption isotherms obtained, four three-parametric modified models of Chung-Pfost, Halsey, Oswin, and Henderson were chosen::

Modified Chung-Pfost

$$a_{w} = exp\left[\frac{-A}{t+B}\exp(-CM)\right] \tag{1}$$

Modified Halsey

$$a_w = exp\left[\frac{-\exp\left(A + Bt\right)}{M^c}\right] \tag{2}$$

Modified Oswin

$$M = (A + Bt) \left(\frac{a_w}{1 - a_w}\right)^c \tag{3}$$

Modified Henderson  

$$1 - a_w = exp[-A(t + B)M^c]$$
, (4)

where: M is the average moisture content, % d.b.;  $a_w$  is the water activity, decimal; A, B and C are coefficients; t is the temperature, °C.

In order to choose the most appropriate of the four models, we applied the eligibility criteria, namely the mean relative error P (%), the standard error of moisture (SEM), and the randomness of residuals:

$$P = \frac{100}{N} \sum \left| \frac{M_i - \dot{M}_i}{M_i} \right| \tag{5}$$

$$SEM = \sqrt{\frac{\sum (M_i - \hat{M}_i)^2}{df}}$$
(6)

$$e_i = M_i - \hat{M}_i \quad , \tag{7}$$

where:  $M_i$  and  $\hat{M}_i$  are experimentally observed and predicted by the model value of the equilibrium moisture content; Nis the number of data points; A, B and C are coefficients; dfis the number of degree of freedom (number of data points minus number of constants in the model).

To calculate the monolayer moisture values, we linearized the Brunauer-Emmett-Teller (BET) model (Brunauer et al., 1938):

$$M = \frac{M_{e}Ca_{w}}{(1 - a_{w})(1 - a_{w} + Ca_{w})} \quad , \tag{8}$$

where: *M* is the MMC, % d.b.;  $a_w$  is the water activity, decimal; *C* is the coefficient.

A detailed method description and estimation of sorption characteristics are reported by Bogoeva (2020). All reagents used in the study were of analytical grade. All tests were conducted in triplicate runs. The data presented are mean values and standard deviations.

However, we were not able to find any information concerning an examination of a flour mixture that consists of these two regional products – oat flour and nectarine powder, intended for sponge cake production. This gives us a reason to conduct the present research.

### **Results and Discussion**

# Physicochemical parameters, antioxidant activity and microbiological load

The first steps in researching the new flour mixture of oat flour enriched with nectarines powder were to determine the general parameters, which were as follows:: total lipids 3.71±0.72 g, protein 9.98±0.07 g, total carbohydrates 74.40±0.86 g, crude fibers 6.21±0.10 g and ash 3.49±0.19. These results were calculated on a dry matter basis: 90.82±0.18%. The mixture was characterised by a lower protein content compared to pure oat flour due to the very low protein content of nectarine powder. Oats are classified as one of the most nutritious grain cereals due to their high protein and fiber content and their high amounts of minerals and vitamins (Youssef et al., 2016; Ahmad & Zaffar, 2014). The interest in oat products has increased in recent years for the sake of high soluble dietary fiber ( $\beta$ -glucan) that has been shown to reduce cholesterol levels and modulate gut microbiota in humans (Joyce et al., 2019).

An *in vitro* study was conducted to prove the fundamental content of bioactive components, it found the presence of antioxidants. Four methods, which differ in mechanism and mode of action, were used. Before analysis of antioxidant activity, the triple extraction of the sample was performed with 70% ethanol. The yield by extraction of a 1 g sample was 26.97%. The results are shown in Table 1, expressed in mMTE/g extract and mMTE/g dry matter basis.

Oats are rich in phenolic compounds such as flavonoids, phenolic acids, and avenanthramides, which possess high

 Table 1. Antioxidant activity of oat flour enriched with nectarine powder

Method	mMTE/g	mMTE/g
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	extract	dry matter basis
DPPH	$20.49 \pm 0.09$	5.53±0.04
ABTS	$1.96{\pm}0.05$	0.53±0.01
FRAP	$1.15 \pm 0.04$	0.31±0.01
CUPRAC	$4.60{\pm}0.04$	1.24±0.01

antioxidant activity (Zhang et al., 2021). In addition, oats are well tolerated by many celiac disease patients, for whom nectarine-enriched mixture would be a healthier alternative to plain oat flour due to higher antioxidant capacity and Bvitamin content (Ahola et al., 2020; Mitic et al., 2016). The present study is about flour mixture and more studies are needed on the effect of nectarine powder addition on dough formation, rheology, and organoleptics.

The microbiological safety of the product was ensured and presented in Table 2 by examining the following parameters: the total count of mesophilic aerobic and facultative anaerobic microorganisms, yeasts and molds, *Escherichia coli*, *Salmonella* spp., coagulase-positive staphylococci, and coliforms.

The examined parameters – total number of mesophilic aerobic and facultative anaerobic microorganisms, molds and yeasts, and coliforms are under permissible norms, corresponding to the standard requirements. The presented data confirms no living pathogenic flora was detected on the investigated enriched flour mixtures. From the microbiological analysis, it can be concluded that the flour mixture intended for sponge cakes was characterized by high quality and safety.

# Moisture Sorption Analysis of flour mixture – oat flour with nectarine powder from Bulgarian origin

Before starting the sorption analysis, the initial moisture content of the flour mixture was  $10.79\pm0.16\%$  d. b. Preparation for the sorption analysis included hydration (moisturizing) and dehydration (drying) of the sample for 10 days over distilled water and respectively over  $P_2O_5$  in desiccators before its placement in hygrostats. Following this, the sample was evaluated under various conditions (temperatures and water activities) for 30 days or within the period for achieving the equilibrium moisture content. The moisture content after drying was  $9.41\pm0.07\%$  d. b. for the adsorption process and after moisturizing was  $18.84\%\pm0.12$  d. b. for the desorption moisture content.

Table 2. Microbiological parameters of new flour mixture of oat flour enriched with nectarine powder

Total number of mesophilic aerobic and facultative anaero-	Yeasts and molds , CFU/g	Coliforms, CFU/g	Coagulase-positive staphylococci,	<i>Escherichia coli</i> , CFU/g	Salmonella spp.,/25 g
bic microorganisms, CFU/g			CFU/g		
5.0×10 <sup>4</sup>	8.5×10 <sup>2</sup>	6.7×10 <sup>3</sup>	<10	<10	Absent

Table 3. Equilibrium Moisture Content (EMC) and Standard deviation (SD) of oat flour enriched with nectarine powder of Bulgarian origin for adsorption process

		10°C			25°C			40°C	
Sel	a <sub>w</sub>	EMC	SD	a <sub>w</sub>	EMC	SD	a <sub>w</sub>	EMC	SD
LiCl	0.113	10.16	0.12	0.113	9.57	0.17	0.112	7.90	0.04
CH <sub>3</sub> COOK	0.234	11.87	0.12	0.225	10.82	0.04	0.201	10.39	0.01
MgCl <sub>2</sub>	0.335	14.20	0.05	0.328	12.24	0.08	0.316	12.10	0.13
K <sub>2</sub> CO <sub>3</sub>	0.431	16.47	0.08	0.432	15.55	0.02	0.432	13.90	0.12
MgNO <sub>3</sub>	0.574	18.88	0.06	0.529	18.31	0.03	0.484	16.10	0.17
NaBr	0.622	20.90	0.01	0.576	19.44	0.12	0.532	18.14	0.11
NaCl	0.757	28.15	0.08	0.753	25.87	0.05	0.747	25.07	0.09
KCl	0.868	36.56	0.13	0.843	34.39	0.08	0.823	31.41	0.17

Table 4. Equilibrium Moisture Content (EMC) and Standard deviation (SD) of oat flour enriched with nectarine powder of Bulgarian origin for desorption process

		10°C			25°C			40°C	
Sel	a <sub>w</sub>	EMC	SD	a <sub>w</sub>	EMC	SD	a <sub>w</sub>	EMC	SD
LiCl	0.113	9.36	0.08	0.113	7.90	0.09	0.112	7.39	0.11
CH <sub>3</sub> COOK	0.234	13.46	0.15	0.225	12.08	0.16	0.201	9.72	0.14
MgCl <sub>2</sub>	0.335	15.14	0.12	0.328	14.27	0.11	0.316	10.21	0.08
K <sub>2</sub> CO <sub>3</sub>	0.431	16.85	0.02	0.432	16.56	0.02	0.432	12.29	0.02
MgNO <sub>3</sub>	0.574	17.80	0.07	0.529	17.65	0.13	0.484	12.77	0.12
NaBr	0.622	19.30	0.08	0.576	18.99	0.16	0.532	14.86	0.03
NaCl	0.757	27.86	0.17	0.753	26.99	0.15	0.747	24.47	0.16
KCl	0.868	33.79	0.01	0.843	30.89	0.08	0.823	29.28	0.03

tion process. It was selected at three different temperatures  $-10^{\circ}$ C, 25°C and 40°C for examination of the processes. Table 3 and Table 4 present the equilibrium moisture content, expressed in % d.b. for the adsorption and the desorption processes at 10°C, 25°C and 40°C.

According to the presented data, it could be concluded that the well-known tendency of most powdered food – the equilibrium moisture content decrease when the temperature increase in the condition of constant water activity. This statement is reported also in the research of Bejar et al. (2012), Dalgıç et al. (2012), Hidar et al. (2018), Bogoeva & Durakova (2020), Bogoeva (2020) and Rosa et al. (2021). By studying the sorption data, it is possible to control and prevent some chemical, physical, and organoleptic changes in the product. For example, if the product is stored at 25°C with the relative air humidity in the range of 45% to 55%, the moisture content of the product has to be maintained from 15.55% to 18.31%.



Fig. 1. Sorption isotherms at 10°C of oat flour enriched with nectarine powder of Bulgarian origin

The classification of the obtained isotherms was investigated in Figure 1 – compared isotherms at  $10^{\circ}$ C.

The graphical analysis demonstrated an *S*-sheped isotherm, therefore the adsorption and desorption isotherms are from the second class, representative of Brunauer's classification (Brunauer et al., 1940). These isotherms are distinctive of the numerous floury food products and are also reported in the studies of Bejar et al. (2012), Rosa et al. (2021), Panyathitipong et al. (2022) and Bogoeva & Durakova (2020).

We have opted for four different three-parametrical modified models for fit using the software Statistica (non-linear estimation) to describe the six obtained isotherms. The obtained model coefficients (A, B and C), as well as the results of the applied criteria for evaluation of the models – mean relative error (P, %), standard error of moisture (SEM) and the distribution of the residuals, are presented in Table 5 for the adsorption process and in Table 6 for the desorption process.

According to the principle, the values of  $P \le 10$  and SEM  $\le 10$ , we can offer for both sorption processes the models of Oswin, Halsey, and Chung-Pfost for the description of the isotherms (Bejar et al., 2012; Bogoeva & Durakova, 2020; Bogoeva, 2020). The modified Oswin was carefully chosen as the model with the lowest value and as the most suitable model for the description of both processes. However, a graphical analysis of the distribution of residues shows that the modified Henderson model has a random distribution of residues in both processes. Fortunately, we can summarize that due to the two criteria P and SEM – we recommend the model of Oswin, and according to the distribution of residues – Henderson (Figures 2 and 3).

Taking into account the equilibrium moisture content and the results under  $a_w \leq 0.5$ , we linearized the BET model for predicting the monolayer moisture content values for all

Table 5. Adsorption: model coefficients, P, SEM and distribution of residuals for modified oswin, modified halsey, modified henderson and modified chung-pfost

Model	A	В	С	Р	SEM	Residuals
Oswin	18.144	-0.028	0.382	4.78	0.95	Non-random
Halsey	5.0132	-0.004	1.870	4.48	1.20	Non-random
Henderson	0.00012	11.420	1.779	18.44	4.21	Random
Chung-Pfost	1355.124	0.110	250.095	6.88	1.77	Non-random

Table 6. Desorption: model coefficients, P, SEM and distribution of residuals for modified oswin, modified halsey, modified henderson and modified chung-pfost

Model	A	В	С	Р	SEM	Residuals
Oswin	18.553	-0.068	0.360	7.55	1.51	Non-random
Halsey	5.3870	-0.016	1.927	8.23	2.29	Non-random
Henderson	0.00016	2.872	1.826	20.46	5.18	Random
Chung-Pfost	421.176	0.116	60.180	9.15	1.73	Non-random



\*Residues lager that ±2 are not presented. Fig. 2. Distribution of residues for the adsorption process



\*Residues lager that  $\pm 2$  are not presented.

Fig. 3. Distribution of residues for the desorption process



Fig. 4. Linearization of BET model for the adsorption process

three chosen temperatures  $-10^{\circ}$ C,  $25^{\circ}$ C and  $40^{\circ}$ C. The linearization of the BET equation and obtained coefficients were demonstrated in Figure 4 for the adsorption and desorption process (Brunauer et al., 1940; Dalgıç et al., 2012; Panyathitipong et al., 2022).

The calculation of the monolayer moisture content was performed using the coefficient of the equation BET. The results are presented in Table 7 for the two sorption processes of adsorption and desorption and temperatures  $-10^{\circ}$ C, 25°C and 40°C.

 Table 7. Monolayer moisture content, % d.b. of oat flour

 enriched with nectarine powder of Bulgarian origin

Process of	MMC, % d. b.					
	10°C	25°C	40°C			
Adsorption	8.98	9.17	8.18			
Desorption	10.02	10.19	6.93			

The results for monolayer moisture content show that the temperature dependence for increasing and decreasing the equilibrium moisture content is not valid here. These are special cases that are also found in the studies of Dalgiç et al. (2012), Lam et al. (2012), Bogoeva (2020) and Bogoeva & Durakova (2020). Therefore, each product has its own specific sorption characteristics, which must be studied before the stages of processing, drying, storage, and transportation.

#### Conclusions

A flour mixture of oat flour and nectarine powder of Bulgarian origin was investigated. Nutritional values of the sample were total lipids 3.71g, protein 9.98 g, total carbohydrates 74.40 g, crude fibers 6.21 g and ash 3.49 g, calculated on a dry matter basis of 90.82%. The content of bioactive compounds was proved using the *in vitro* methods DPPH, ABTS, FRAP, and CUPRAC and the results were presented in mMTE/g extract and mMTE/g d.b. The microbiological safety of the product was guaranteed by analyzing the following parameters: the total count of mesophilic aerobic and facultative anaerobic microorganisms, molds and yeasts, *Escherichia coli*, *Salmonella* spp., coagulase-positive staphylococci, and coliforms.

The results showed that no living cells or pathogenic microorganisms were detected. The sorption characteristics - equilibrium moisture content and monolayer moisture content were determined at three temperatures - 10°C, 25°C and 40°C for eight different water activities in the 0.1 - 0.9range. This research confirmed that when the temperature increase, the equilibrium moisture content decrease in the condition of constant water activity. A graphical analysis of sorption isotherms demonstrated the S-shape profile character for the second class of Brunauer's classification. Using the criteria of mean relative error (P, %) and standard error of moisture (SEM), we recommend the modified model of Oswin for a satisfactory description of the sorption capacity. Furthermore, the graphical criteria distribution of the residuals was shown in the modified Henderson for the most random and described isotherms model. The BET equation was linearized for the obtention of the coefficients for the calculation of the monolayer moisture content. The results for the adsorption process are in the range from 8.18% d.b. to 9.17% d.b. and for the desorption process – from 6.93% d.b. to 10.02% d.b.

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