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PHYSICOCHEMICAL CHARACTERICS OF SUGAR BEET MOLASSES USED AS THE MEDIUM FOR OSMOTIC DEHYDRATION OF PORK MEAT

FIZIČKOHEMIJSKE KARAKTERISTIKE MELASE ŠEĆERNE REPE KORIŠĆENE KAO MEDIJUM ZA OSMOTSKU DEHIDRACIJU SVINJSKOG MESA

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ABSTRACT

Physicochemical properties of the sugar beet molasses (SBM), used as the medium for osmotic dehydration (OD) of pork meat, were determined to improve OD process performances. Macroelements content (Na, K, Ca and Mg) decreased in molasses after OD meat treatment, although rather slightly. Differential scanning calorimetry and modulated differential scanning calorimetry revealed that there was a strict dependence between water content and temperature of molasses glass transition, and suggested the character of the thermal process. Influence of the high organic content of molasses on the secondary lipid oxidation products of OD meat were determined using thiobarbituric acid method.

***Key words:** molasses, osmotic dehydration, macroelements, lipid oxidation, differential scanning calorimetry.*

REZIME

Fizičkohemijske osobine melase šećerne repe, korišćene kao medijum za osmotsku dehidraciju (OD), svinjskog mesa, su bile praćene da bi se popravile karakteristike OD procesa. Sadržaj makroelemenata (Na, K, Ca and Mg) u melasi opada posle OD tretmana mesa, iako dosta slabo. Diferencijalna skanirajuća kalorimetrija i modulirana diferencijalna skanirajuća kalorimetrija su pokazale da postoji stroga zavisnost između sadržaja vode i temperature staklastog prelaza u melasi, i ukazale na karakter termalnog procesa. Uticaj visokog sadržaja organskih materija u melasi na sekundarne oksidativne proizvode lipida u mesu je bio praćen korišćenjem metoda tiobarbituratne kiseline.

***Ključne reči:** melasa, osmotska dehidratacija, makroelementi, oksidacija lipida, diferencijalana skenirajuca kalorimetrija.*

INTRODUCTION

Osmotic dehydration (OD) is one of the most challenging operations in food dehydration, based on partial removal of water from the food material by direct contact of the product with a hypertonic solution of salts, sugars, acids, etc., *(Raoult-Wack, 1994)*. Sugar beet molasses (SBM), a byproduct of sugar manufacture, is used as the medium for osmotic dehydration (OD), primarily due to the high dry matter (65-80%) and specific nutrient content *(Luczyñska et al., 2009)*. Sugars, plant pigments and organic compounds formed during sugar processing, as well as minerals and vitamins from SBM penetrate into the food material. Reduction in moisture and enhancement of macro element content *(Luczyñska et al., 2009; Filipčev et al., 2010; Della Rosa et al., 2001)* in the food matrix are among main parameters followed in osmotic dehydration and quality of analytical techniques is crucial for of improvement of OD process performances. SBM is a very specific material and the selection of the appropriate mineral extraction procedure *(Zlatanovic et al, 2012)*, precise enough for determining the changes of mineral content of SBM, during osmotic dehydration of pork meat, was implemented. As water activity (a_w) alone was proved to be inadequate in some cases, the concept of glass transition was used in last two decades as a parameter for quantifying water mobility and food stability *(Oliveira et al., 1999)*. In this work, thermal characteristics, by the means of differential scanning calorimetry (DSC) and modulated differential scanning calorimetry (MDSC), of molasses, used for osmotic dehydration, has been studied in the aim of better understanding of molasses' glass transition phenomenon. Reduction in moisture and a_w and increase of salts concentration lead to the reduction in microbial growth and differences in lipid oxidation status *(Singhal et al., 1997)*. The thiobarbituric acid (TBA) test is widely used to evaluate secondary lipid oxidation products in meat. Malondialdehyde (MDA), a secondary oxidation product of polyunsaturated fatty acids (PUFA) with three or more double bonds, reacts with TBA to form a stable pink chromophore with maximal absorbance at 532 nm (TBARS532). Certain compounds interfere with the reaction between TBA and MDA: sugars, water-soluble proteins and peptides, DNA, volatile aldehydes different to MDA, pigments, amino acids, nitrites, metals and compounds with similar spectral properties to that of the TBA–MDA adduct. Numerous method modifications were developed to reduce these interfering reactions *(Fernandez et al., 1997; Wang et al., 2002)*. High level of sucrose (44-54%), other sugars and their degradation products in molasses, used in osmotic dehydration of meat, could generate interfering yellow chromogen (at 450-460 nm) overlapping the pink peak (at 530-537 nm) of TBA-MDA adduct. The yellow chromogen is formed by a variety of aldehydic compounds reacting with TBA *(Fernandez et al., 1997)*. In this work characterization of SBM, considering mineral content, thermal analysis and oxidation capacity, i.e. the interferences in the TBA method, caused by the sugar beet molasses used as solution for osmotic dehydration of pork meat, were studied.

MATERIAL AND METHOD

Experimental setup: Preparation of osmotically dehydrated pork meat in sugar beet molasses (SBM) has been described in detail in *(Pavlović et al., 2012)*. Briefly, fresh pork (Musculus brachii) of normal pH (6.05), was cut into approximately 1x1x1 cm (1cm^3) cubes and osmotically dehydrated in solutions of sugar beet molasses (81,95 °Brix) at 22°C for 5 hours. Solution to sample ratio was 10:1 to avoid significant dilution of the medium by water removal, which would lead to the local reduction of the osmotic driving force during process. SBM samples before and after osmotic treatment were analyzed.

Physicochemical analysis: Dry matter and water content of meat and SBM samples was determined at 105 °C in a laboratory oven until constant weight was achieved, according to the ISO 1442:1997. The combination of thermal treatment at 350 °C, and wet acidic treatment at 160 °C was used for samples preparation for determination of mineral content. The dry samples were processed for minerals determination by wet digestion, where ca. 5 g each, were weighed exactly to four decimal places, and transferred to the vessels, into which 4.5 ml 65% HNO3 and 10.5 ml 35% HCl were added. The treatments were repeated to obtain the white sediments that were dissolved in 0.07 M HNO3. The content of metals present in the corresponding solutions was determined by inductively coupled plasma optic emission spectrometry (ICP-OES). ICP-OES measurement was performed using Thermo Scientific ICAP 6500 Duo ICP (Thermo Fisher Scientific, Cambridge, United Kingdom) spectrometer equipped with RACID86 Charge Injector Device (CID) detector, standard glass concentric nebulizer, quartz torch, and alumina injector. SBM samples have been studied by differential scanning calorimetry (DSC) in the cyclic heat-cool-heat scans, in the temperature range from -90 ^{o}C to 90 ^{o}C, heating rate Hr=10 ^{o}C/min and cooling rate Cr=5 ^{o}C/min, N_2 purge flow of 50 ml/min. MDSC cyclic heat-cool-heat scans were conducted in temperature range from -90 ^{o}C to 90 ^{o}C, with heating rate Hr (r_H)=5 ^{o}C/min with modulation of 0.8 ^{o}C amplitude and 60s period of modulation, under N_2 purge flow of 50 ml/min. Moisture was determined by standard method JUS ISO 1442: 1997. All scans were performed on the DSC Q1000 calorimeter**,** TA Instruments, DE, USA. The Universal AnalysisTM software was used to obtain the glass transition parameters (onset, T_{onset}; midpoint, Tg; final T_{end}, and the shift in specific heat capacity ΔCp). TBA test for secondary lipid oxidation products was determined in OD meat and SBM using aqueous acid extraction TBA method (EM), *(Wang et al., 2002)*. The incubation conditions in EM for TBA-reactive substances **(**TBARS) (Fernandez et al., 1997) were: A) boiling temperature/25 min with 20 mM TBA, B) 40^{o}C/70 min, with 80 mM TBA, and C) 20 ^{o}C/20 h with 20 mM TBA. The steam distillation TBA method (DM) was employed to evaluate the interferences with the spectrophotometric measurement at around 532 nm found in EM. TBARS values were calculated by multiplying the absorbance values at 532 nm by a constant coefficient K_{med}. K_{med} value was derived from standard curves and known dilutions of MDA standard (1,1,3,3 tetrametoxypropane, TMP), and expressed as mg MDA/kg of sample.

Statistical analysis: Microsoft Excel software (Microsoft Office 2003) was used for statistical analysis. All measurements were performed in triplicate.

RESULTS AND DISCUSSION

Several samples of sugar beet molasses (SBM), obtained from three Serbian sugar mills with different water content, were used for osmotic dehydratation of pork meat. Treatment of the pork meat in molasses resulted in the enrichment of meat with minerals and the reduction of mineral content of SBM. Contents of macroelements: Na, K, Ca and Mg in SBM before and after osmotic dehydration, are shown in Table 1.

Table 1. The content of macro elements in sugar beet molasses (SBM), determined by ICP-OES. Results represent mean value ± standard deviation.

Sample	Dry matter at 105 ^{o}C (%)	Moisture at 105 ^{o}C (%)	Na (mg/100 g)	K (mg/100 g)	Ca (mg/100 g)	Mg (mg/100 g)
1. SBM before OD	79.2±0.6	20.7±0.6	749±21	1939±9	289±3	38±1
2. SBM after OD	73.2±0.8	26.8±0.8	689±7	1731±12	268±6	36±1

SBM is a very specific material and the selection of the appropriate mineral extraction procedure is not an easy task. Differences in sample preparation, selection and implementation of the procedure as well as inhomogeneity of the samples were recognized as the main reasons for results variation. The results obtained, with relative standard deviations below 5%, enable reliable observation of changes in the macro elements composition in all materials during the OD process. Comparison of macro elements content between SBM (before OD) and after OD, had shown significantly higher levels of Na, K and Ca in SBM, which was not the case with Mg content. OD pork meat had a significant increase in the content of Na, K and Ca cations with respect to the content obtained for the raw pork. The content of Mg cation in the OD pork meat was slightly raised and reaches a value similar to the SBM *(Zlatanović et al., 2012)*. The content of macro elements (Na, K and Ca) in SBM before and after OD, differs slightly, but significantly, despite the large ratio of OD solution (SBM) to meat samples. Analysis of the mineral content of materials during the process of osmotic dehydration allows better control of better control of the technological process.

From the results obtained, it was found that there was a strict dependence between water content and molasses glass transition (Tg) temperature what is in agreement with literature data (*Sopade, 2007)*. The transition temperatures (midpoint), Tg, ranged from -54.3^{o}C to -32.1^{o}C, while the change in heat capacity (ΔCp) was from 1.1 to 2.4 J/g/^{o}C (Fig. 1. and Table 2.). Dried molasses Tg was found to be -33.0 ^{o}C, which is in agreement with the results obtained by the Fox model for glass transition prediction and predicted Tg of the anhydrous molasses *(Sopade et al., 2007)*. The samples with the highest moisture showed the lowest glass transition temperatures, while the highest glass transition temperatures were obtained from the most dry molasses samples. Using MDSC, the reversing and nonreversing thermal events in the low temperature region of molasses have been obtained, suggesting on the existence of only one thermal process.

Table 2. Glass transition temperature (Tg), heat capacity (ΔCp) and moisture (%) of sugar beet molasses (SBM) and dried SBM.

No.	Sample	Tg (^{o}C)	Δ Cp(W/g)	Moisture (%)
1.	SBM before OD	-40.04	0.1651	17.66
2.	SBM 3. after OD	-33.54	0.1374	28.03
3.	SBM before OD	-42.20	0.1571	20.70
4.	SBM 5. after OD	-38.51	0.1558	26.79
5.	SBM before OD	-47.81	0.1848	20.73
6.	Dried SBM	-32.53	0.0823	0.00

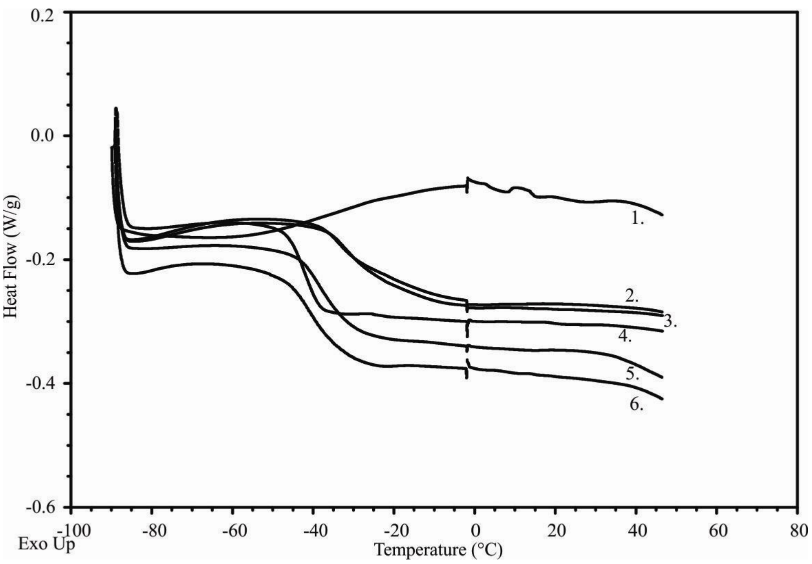

Fig. 1. MDSC curves of sugar beet molasses (SBM) before and after osmotic dehydration process and of dried SBM. SBM samples 1-6 are designated as in Table 2

Interferences, which cause erroneously high value of TBARS in meat samples dehydrated in molasses, were detected as absorption at 350 nm, 450-460 nm, and 532 nm in SBM itself, using aqueous acid extraction method (EM) under different conditions of incubation (A, B and C) (Pavlović et al., 2012). These spectrophotometric interferences, found in diluted molasses solutions, are overlapping the pink peak (532 nm), characteristic for MDA. The reaction with TBA was dependent on molasses concentration and was maximal at 100^{o}C, as shown in Fig. 2. (scan 2 and 4).

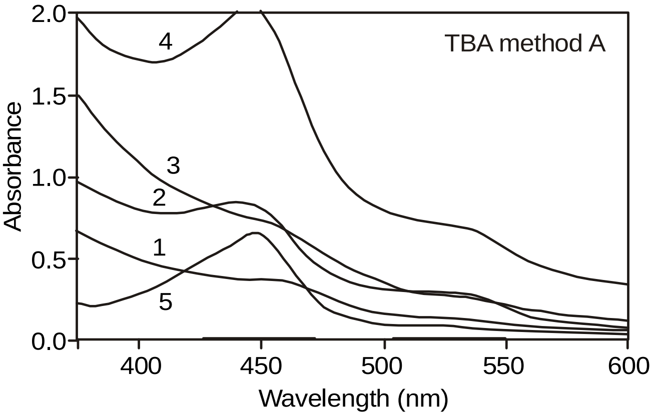

Fig. 2. Scans of sugar beet molasses and sucrose absorbance in TBA method A (EM, 100^{o}C/25 min, 20 mM TBA). Scans: 1) 10% molasses, before TBA reaction; 2) 10% molasses, after TBA reaction; 3) 20% molasses, before TBA reaction; 4) 20% molasses, after TBA reaction, 5) 10% sucrose after TBA reaction

It was slower when the incubation temperature was decreased, (data not shown). It is known that sucrose reacts with TBA, generating a yellow adduct with maximum absorbance at 450 nm (Fig. 2., scan 5), when the incubation temperature is above 50 °C *(Wang et al., 2002)*. It is evident that some other TBA-reactive compounds, possibly antioxidants *(Guimaraes et al., 2007)*, as melanoidin compounds and caramels, may be present in the sugar molasses, which can lead to interferences with the spectrophotometric measurement and hence, cause an overestimation of the results. The reaction between TBA and MDA is more specific at room temperature than at boiling temperature. Hence, the interfering colorings can be reduced by incubating TBA and MDA at lower temperatures, while the reaction time can be reduced by increasing of TBA concentrations from 20 mM to 80 mM. The modified method can be used to measure MDA in the presence of high concentrations of interfering sucrose (higher than 4%), or other similar interfering agents *(Wang et al., 2002)*. The method of Wang et al. (TBA method B) could be the appropriate method for TBARS measurement in pork meat osmotically dehydrated in SBM which contained 4-13% of sucrose. The difficulty to determine the optimal conditions for the release of MDA from its bound forms in muscles without using strong acidic conditions and heating, which, could destabilize the MDA-TBA complex *(Fernandez et al., 1997)*, was suggestive of combining mild extraction procedure *(Wang et al., 2002)* with different incubation temperatures and TBA concentrations in EM. No significant differences in TBARS values were found in molasses samples before and after OD.

CONCLUSION

Sugar beet molasses (SBM), a byproduct of sugar manufacture, used as the medium for osmotic dehydration, is a very specific material in terms of the selection of the appropriate methods for monitoring of the OD process. The combination of thermal treatment at 350 ^{0}C, and wet acidic treatment at 160 ^{o}C was found as optimal for the mineral extraction procedure. Content of macro-elements (Na, K, Ca and Mg) decreased in molasses after OD meat treatment, although rather weakly. Differential scanning calorimetry and modulated differential scanning calorimetry revealed that there was a strict dependence between water content and temperature of molasses glass transition. Samples of highest moisture showed the lowest glass transition temperatures, while the highest glass transition temperatures were obtained with the sample of dried molasses. Using MDSC, the reversing and non reversing thermal events in the low temperature region of molasses have been obtained, suggesting on one thermal process. The effectiveness of three TBA tests in minimizing the interferences of molasses ingredients was measured by aqueous acid extraction method (EM) under different conditions of incubation. TBA-EM at high incubation temperature (100^{o}C) may be an inadequate method for the analysis of oxidative deterioration of meat samples dehydrated in molasses, due to interferences which cause erroneously high values of TBA, detected as absorption at 350 nm and 450-460 nm, which are overlapping the pink peak (max. 532 nm), characteristic for MDA. TBA-EM procedure at low incubation temperature (40^{o}C) and elevated TBA concentration (80 mM) was the most sensitive of the studied TBA methods.

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REFERENCES

Della Rosa, M., Giroux, F. (2001). Osmotic treatments (OT) and problems related to the solution management, J. Food Eng., 49, 223-236.

Fernandez, J., Perez-Alvarez, J. A. & Fernandez-Lopez, J. A. (1997). Thiobarbituric acid test for monitoring lipid oxidation in meat, *Food Chemistry*, 59 (3), 345-353.

Filipčev, B., Lević, Lj., Bodroža-Solarov, Marija, Mišljenović, Nevena, Koprivica, Gordana, (2010). Quality Characteristics and Antioxidant Properties of Breads Supplemented with Sugar Beet Molasses-Based Ingredients, Int. J. Food Prop., 13, 1035-1053.

Guimaraes, C. M. , Giao, M. S. , Martinez, S. S. , Pintado, A. I. , Pintado, M. E. , Benito, L. S. and Malcata F. X. (1997). Antioxidant Activity of Sugar Molasses, Including Protective Effect Against DNA Oxidative Damage, *Journal of Food Science, 72*, 1, C39 - C43.

ISO 1442/1997: Meat and meat products - Determination of moisture content (Reference method)

Luczyñska, J., Toñska, E., Luczyñski, M. J. (2009). Essential mineral components in the muscles of six freshwater fish from the Mazurian great lakes (Northeastern Poland), Arch. Pol. Fish., 17, 171-178.

Oliveira, J. C., Pereira, P. M., Frias, J. M., Cruz, I. B., MacInnes, W. M. (1999). Application of the concepts of biomaterials science to the quality optimization of frozen foods. In F. A. R. Oliveira, & J. C. Oliveira (Eds.), Processing foods quality optimization and process assessment, 107–130). USA: CRC Press LLC

Pavlovic, M., Mitić-Ćulafić, D., Mandić, M., Šuput, D., Ostojić, S., Pezo, L., Lević, Lj. (2012). Suitability of thiobarbituric acid method for assesing lipid oxidation in pork, osmotically dehydrated in sugar beet molasses Proceedings of 6th Central European Congress ON Food, 23-26 May 2012, Novi Sad, Serbia, 2, 507-512

Raoult-Wack, A.L. (1994). Recent advances in the osmotic dehydration of foods. Trends in Food Science & Technology 5, 255–260.

Singhal, R.S., Kulkarni, P.K., and Rege, D. V. (1997). Handbook of indices of food quality and authenticity, Woodhead Publishing, Cambridge, UK.

Sopade, P. A., Lee, S. B., Whitea, E. T., Halley, P.J. (2007). Glass transition phenomena in molasses. LWT - Food science and technology 40, 1117–1122.

Wang, B., Pace, R.D., Dessai, A.P., Bovell-Benjamin, A., Phillips, B. (2002). Modified Extraction Method for Determining 2-Thiobarbituric Acid Values in Meat with Increased Specificity and Simplicity. *Journal of Food Science, 67*, 8, 2833-2836.

Zlatanović, S., Kovaćević, O. A., Ostojić,S., Dojčinović, B. P., Pavlović, M., Šuput. D. (2012). Macro elements composition of the osmotically dehydrated pork meat in sugar beet molasses solution, Proceedings of the 11th internationalconference of fundamental and applied aspects of physical chemistry - PHYSICAL CHEMISTRY 2012, September 24-28, Belgrade, Serbia, 2, 766-768.

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