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Effect of Freeze-Dried Celery Products on the Glutamic Acid Content in Model Meat Systems Under Different Ripening Conditions

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Abstract. The effect of celery products (3 %), starter culture and ripening conditions on pH and free glutamic acid content in model meat system were evaluated. For that reason model meat system from minced pork, lyophilized celery products and starter cultures were formulated and ripened at different conditions. It was determinated that carbohydrates presented in celery products and higher temperature (20 – 24 ^{o}C) influenced the faster decrease of pH in model meat system. Ripening process for 10 hours at +8 ^{o}C was too short for protein degradation and free glutamic acid formation. Due to the action of starter culture and endogenous meat enzymes free glutamic acid content increased in 4 – 5 times after 4 days of ripening at 20 – 24 ^{o}C temperature. Added freeze-dried celery products did not affect glutamic acid content.

Keywords: ripening, glutamic acid, celery, model meat system

1. Introduction

It is predicted that by 2020, meat consumption will constantly increase. Manufacturers are trying to create a healthier meat products therefore are looking for new technological solutions. One of the options is the replacement of synthetic additives with natural plant origin ones, which have similar functional and technological characteristics as synthetic. Celery (*Apium graveolens*) is widely used in human nutrition. This vegetable is rich in vitamins, minerals, essential oils and active substances such as polyphenols. It is also known as good source of nitrates [1]. In addition to these components celery contains free glutamic acid. L-Glutamic acid (Glu) is an amino acid presented in foodstuffs in the free and protein-bound forms. In its L-configuration, free Glu acts as flavour enhancing agent and is widely used in food industry, particularly in the form of the monosodium salt (MSD). It produces a unique taste that cannot be provided by other basic taste (saltiness, sourness, sweetness and bitterness), referred to as a fifth taste (umami) [2]. There is no complete agreement about the safety of MSG, even though the Food and Drug Administration (FDA) includes it among the substances generally recognized as safe (GRAS) [3]. The optimum amount of added MSD to enhance the taste of food is at 0.1–0.8 % by weight. For instance, food of 500 g needs 0.5–4.0 g of glutamate to bring a good taste, which is the same as that of glutamate naturally found in general food. For example, protein from meat contains 11–22 % of glutamate, whereas plant protein has about 40 % of glutamate [4].

Drying is an ancient process used to preserve foods. The main purpose of drying is to remove water and stop enzymes activity. While selecting the method of drying and its parameters it is important to consider the fact that most of vitamins and other biologically active substances are lost during the process due to their sensitivity to heat and enzymatic oxidation. In order to preserve the nutritional and biological value of vegetables they can be freeze – dried. Freeze – drying is the process based on the dehydration by sublimation of a frozen product. Due to the absence of liquid water and the low temperatures required for the process, most of the deterioration and microbiological reactions are stopped resulting a final product of excellent

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quality. Lyophilization compared to other drying methods has many advantages, including good sensoric properties and high nutritional value [5].

The objective of this study was to investigate the effect of freeze-dried celery products on the free glutamic acid content in the model meat systems at different ripening conditions and starter cultures added to the system.

2. Materials and Methods

2.1. Celery Products

Five different celery cultivars (*Grone pascal*, *Malachit*, *Giant Pascal*, *Hellios* and *Zefir*) from the experimental garden of Institute of Horticulture of Lithuanian Research Centre for Agriculture and Forestry were for the manufacture of two freeze-dried celery products: lyophilized celery (LC) and lyophilized water soluble substances of celery (LWSC). For the production of LC celery stalks were washed, chopped, frozen at minus 18 ^{o}C and freeze dried using sublimator Zirbus 4x5x6 (Germany). Lyophilized celery stalks were crushed to powder-like state and then sieved through 1.5 - 2.0 mm sieve. For the production of LWSC celery juices were obtained from the celery stalks which were washed and pressed using the low-speed press. Pomarce was mixed with water and stirred periodically during 3 h at 20 ^{o}C temperature. Celery juice and water extract were mixed, frozen at minus 18 ^{o}C and freeze dried using sublimator Zirbus 4x5x6 (Germany).

2.2. Model Meat System Formulation and Processing

Model meat systems were composed from minced pork meat with the addition of 3 % freeze-dried celery products, spices and starter culture. Formulation of model meat systems is shown in Table I. Salt and celery products concentrations are approximately those used in the sausages. After mixing all ingredients the ball shape samples of model meat systems were hand formed (each sample weighted approximately 30 $\pm$ 1 g and was 50 $\pm$ 2 mm in diameter). Model meat systems were ripened under two different conditions: 10 hours in refrigerator at 8 ^{o}C temperature; 4 days in refrigerated incubator *Ing* Climas CIR 322/HR (Spain) by lowering the temperature and relative humidity from 24 ^{o}C, 92 % to 20 ^{o}C, 88 %, respectively.

Table I: Formulation for model meat system (10.0 kg) with 3 % celery products additive

Ingredients	*Control*	*Control with starter culture*	*Model meat system with LC*	*Model meat system with LWSC*	*Model meat system with LC and starter culture*	*Model meat system with LWSC and starter culture*
The main materials, kg						
Pork meat	7.95	7.95	7.95	7.95	7.95	7.95
Pork backfat	1.75	1.75	1.75	1.75	1.75	1.75
LC	-	-	0.3	-	0.3	-
LWSC	-	-	-	0.3	-	0.3
Spice, g/kg						
Salt	27	27	27	27	27	27
Black pepper	2	2	2	2	2	2
Coriander	1	1	1	1	1	1
Starter culture, g/kg						
St. xylosus; St. carnosus; Pediococcus pentosaceus	-	1.6	-	-	1.6	1.6

2.3. Compositional Analysis

The chemical composition of freeze-dried celery products was evaluated by moisture, protein, fat and ash content which were measured following stadard methods. Moisture content was determined by drying at 105 ℃ to the constant weight. Nitrogen content was determined by the Kjeldahl method and proteins were estimated by multiplying the nitrogen content by 6.25. Total lipids were extracted from the sample using chloroform as solvent. Total mineral content was determined by drying at 500-600 ^{o}C to a constant weight. The carbohydrates content was calculated.

2.4. pH

The pH was measured with a pH meter (WTW 3110 GmbH, Germany) equipped with a pH probe (N1048 A, Schott Instruments) calibrated with standard buffers at pH 4.0 and pH 7.0. The pH was

determined by inserting the probe directly into the model meat system. For each treatment, measurements were made in triplicate. Measurements were carried out at 20 ℃.

2.5. L – Glutamic Acid

Glutamic acid content was determinated by means of BIOEHRINGER MANNHEIM/ R – BIOPHARM enzymatic kit (Roche, Germany). For celery products: 1 g of sample was extracted with 50 ml of water (for 10 min) and transferred to the 100 ml volumetric flask, filled up to the mark with water, shaked and filtered. For model meat system: 10 g of minced meat sample was homogenized with 80 ml perchloric acid (1M) for 10 min; the supernatant was decanted and filtered. 20 ml of filtrate was pipetted into the breaker and pH was adjusted to pH 10.0 with KOH (2 M); the volume of KOH was registered. To obtain the quantitative precipitation of the potassium perchlorate formed, the beaker was placed into an ice-bath for 20 min and filtered. The clear solution dilute was used for the assay. Following the description of BIOEHRINGER MANNHEIM/ R – BIOPHARM enzymatic kit L – Glutamic acid is oxidatively deaminated by nicotinamideadenine dinucleotide (NAD) to 2 – oxoglutarate in the presence of the enzyme glutamate dehydrogenase. In the reaction catalyzed by diaphorase the NADH formed converts iodonitrotetrazolium chloride (INT) to a formazan which is measured at its maximum in the visible range at 492 nm. The equilibrium of reaction lies on the side of L – glutamate. By trapping the formed NADH with INT, the equilibrium is displaced in favour of 2 – oxoglutarate.

3. Results and Discussion

3.1. Chemical Composition of Celery Products

The analysis of chemical composition of celery products showed that LWSC contained 1.37 times higher moisture (7.73 %), 1.5 times higher protein (7.23 %) and 4.58 % lower carbohydrates (74.74 %) amount in comparison with LC (Table II.). The mineral content was similar in both products and was in the range 9.15 – 9.76 %.

It was found that free glutamic acid content was 1.87 times higher in LWSC (346.25 mg/100g), compared with that in LC. According to data presented in literature free glutamic acid content in plants depends on the cultivars, cultivation method, harvesting time, the growth conditions, fertilizer type and other factors [6]. The difference in glutamic acid content in celery products could be caused by its ability to dissolve in water [7].

Table II: Chemical composition of celery products

Celery products	Moisture, %	Protein, %	Fat, %	Minerals, %	Carbohydrate, %	Free glutamic acid, mg/100 g (dm)
LC	5,61±0,07	4,81±0,00	1,11±0,35	9,15±0,03	79,32±3,97	185,27
LWSC	7,73±0,07	7,23±0,16	0,54±0,19	9,76±0,27	74,74±3,74	346,25

3.2. pH

The changes of pH during ripening process of model meat systems produced with different celery products under different ripening conditions were measured. The pH values of all model meat system remained constant over the 10 hours of ripening at + 8^{o}C temperature (Figure 1). At the end of the ripening process a small decrease of pH, ranging from 5.58-5.67 to 5.50-5.56 was recorded for all systems. Such behaviour was caused by the low activity of lactic acid bacteria *Pediococcus pentosaceus* at the ripening temperature [8]. Different chages of pH were recorded in the model meat systems during ripening for 4 days at 20-24 ^{o}C (Figure 2). In the model meat systems produced with celery products pH decreased after 2 days of ripening and at the end of the process pH was 5.22-5.28. The pH value of model meat system with the addition of celery products and starter culture decreased after the first day of ripening. It can be seen that under favorable conditions for the fermentation of celery carbohydrates by lactic acid bacteria a significant drop in pH was recorded (pH 4.94) thus ensuring the safety and stability of meat system [9]. The highest pH value (5.64) was determined in the control model meat system without addition of starter cultures and celery products; in this system no changes of pH was noticed during entire ripening process. Relatively high pH values were recorded in control meat system with the addition of starter cultures and no celery products; in this system the pH value decreased from 5.59 to 5.44 during ripening.

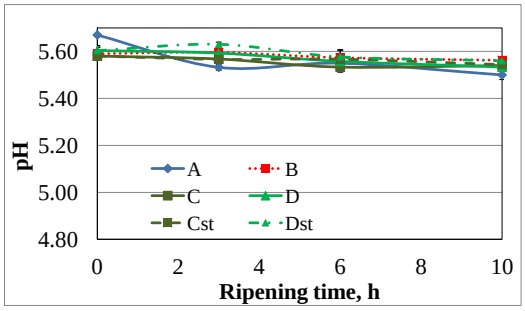

Fig. 1: changes of pH during 10 h ripening of model meat system with celery products:

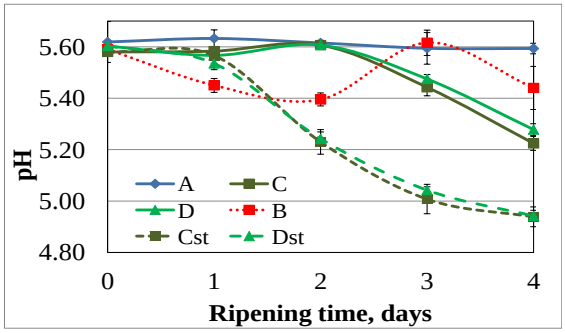

Fig. 2: changes of pH during 4 days ripening of model meat system with celery products:

A – Control; B – Control with starter culture; C – Model meat system with LC; D – Model meat system with LWSC; Cst – Model meat system with LC and starter culture; Dst - Model meat system with LWSC and starter culture

3.3. Free glutamic Acid

The changes in free glutamic acid content in the model meat system produced with starter cultures and different celery products were measured during ripening (Figure 3 and Figure 4). It was found that after 10 h of ripening at 8 °C the glutamic acid content in all model meat systems did not change and was in the range 14.12-15.68 mg/100 g. It is known that very similar amount of glutamic acid is found in meat (9.0-23.0 mg/100 g) without additives [10]. The obtained results indicate that no protein degradation occure in the model meat system at 8 °C because of low activity of the added starter cultures therefore no additional free glutamic acid formation was detected. 8 °C temperature was not favorable for the action of endogenous and bacterial enzymes which play an important role in amino acid generation [11]. Very similar results are reported by Kezban Candogan et al. (2008). They determined that free glutamic acid content in beef sausages produced with starter cultures increased from 61,6 to 74,3 mg/100 g during ripening for 72 h at 8-10 °C [12]. In the case of control model meat system ripened at +24 °C for 4 days the free glutamic acid content increased up to 54 mg/100 g, i.e. was 3 times higher in comparison with that at the beginning of ripening process. Similar results were obtained for the model meat system with LWSC; at the end of ripening process this system contained 53.0 mg/100 g of free glutamic acid. Higher amounts of glutamic acid were recorded after 4 days of ripening of model meat system with the addition of LC (68 mg/100g).

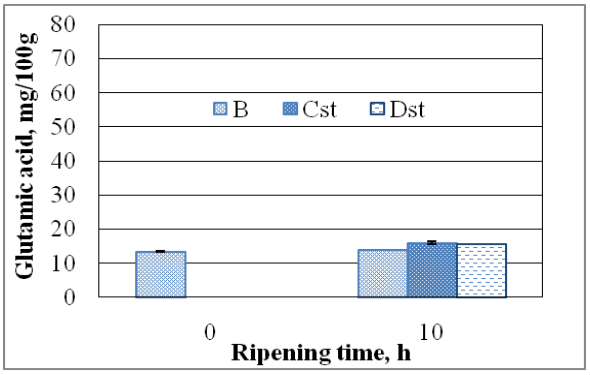

Fig. 3: changes of free glutamic acid during 10 h ripening of model meat system with celery products:

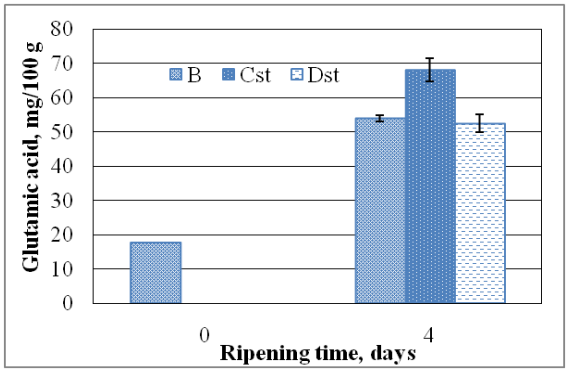

Fig. 4: changes of free glutamic acid during 4 days ripening of model meat system with celery products:

B – Control with starter culture; Cst – Model meat system with LC and starter culture; Dst - Model meat system with LWSC and starter culture

4. Conclusion

Results of our experiment showed that addition of celery products to the model meat system had no negative effect on the ripening process. Carbohydrates presented in celery products, as well as higher temperature and starter culture addition induced the more rapid reduction of pH of meat; that is important for

ensuring the safety and stability of meat product. At ripening temperature 20-24 ^{o}C the glutamic acid content in model meats systems was 4 – 5 higher in comparison with that at 8 ^{o}C. However, the addition of celery products did not increase the free glutamic acid content in model meat systems after ripening.

5. Acknowledgment

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