

USE OF BAR PROCESSING TO INCREASE THE SHELF LIFE OF VITAMINIZED SAUSAGES AND THEIR USE FOR THE CORRECTION OF STUDENTS' HEALTH

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Abstract: One of the priority directions of state policy in the field of healthy food is the development and integration of enriched foodstuffs with an increased expiration date into production. The purpose of researches is the study of effect of bar processing on the periods of storage of boiled sausages enriched with vitamin premix. The test samples of boiled sausages were processed under the pressure of 800 MPa within 3 minutes at a temperature of 0 ... +4°C by means of a hydrostat after the end of the technological process. The control samples of boiled sausages were not processed under pressure. The safety of boiled sausages was estimated by organoleptic and microbiological indicators and pH, the content of vitamins in the product was researched in 8 and 16 days of storage. It has been established that in 16 days of storage the control samples of sausages did not conform to the requirements of regulating documentation; the shift of pH to the alkaline side, the increase in the quantity of mesophilic aerobic and facultative and anaerobic microorganisms has been noted. After 16 days of storage the content of PP and C vitamins in the control samples of sausages authentically decreased by 17.6% and 93.4% while the decrease in the test samples was 4.1% and 12.0%. The antioxidant activity of test samples of sausages is authentically 71.3% higher than that of the control samples (0.12 ± 0.04 mol equiv/dm³). Against a background of use of vitaminized sausages an authentic increase in the antioxidant activity of catalase and ceruloplasmin is noted in the blood of students of the test group. Thus, it is established that the processing of boiled sausages enriched with vitamins, under high pressure has a bactericidal effect on microbic cells, prevents proteolysis, saves vitamins and, respectively, increases the expiration date of a foodstuff. The calculations for design of a high pressure hydrostat for foodstuff processing have been performed.

Keywords: processing under high pressure, meat raw materials, meat products, technical regulations, hydrostat, antioxidants, indicators of quality and safety

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INTRODUCTION

75% of the population of the Russian Federation feel lack of irreplaceable micronutrients in their diet which provides the deterioration of health, a decrease in working capacity, an increase in fatigue and the development of various diseases. One of the etiological factors of hypovitaminoses is a decrease in the quantity of vitamins in foodstuffs due to the use of thermal technologies of processing of food raw materials providing a decrease in a share of biologically active agents in foodstuffs.

In this regard, the development of enriched foodstuffs and the provision of stability of micronutrients in the course of storage is one of the priority directions of the modern food industry. In this case, foodstuff processing under high pressure deserves special attention. The main scope of the method of high pressures in the world today is athermic preservation ("cold pasteurization", pascalization) of foodstuffs aimed at the inactivation of microorganisms and enzymes of the processed environment. In the 90-ies of the last century in Japan the first wave of popularity of

jams of strawberry, kiwi and apples received by the use of high hydrostatic pressure began, in 1997 barotechnology was first used by the company Fresherized Foods – a world leader in the production of Guacamole (a traditional Mexican snack of avocado pulp), in 2007 approximately 120 barometric plants were put into operation for production of "new" products commercially [1] worldwide. More than 80% of the equipment functioning today were collected and produced after 2000 which testifies that this trend tends to the accelerated development and expansion of its scope [2]. Today the following countries dominate in this trend of production: North America (USA, Canada, Mexico); Europe (Spain, Italy, Portugal, France, Great Britain, Germany); Asia (Japan, China, North Korea); Australia.

The total of the products processed under pressure is steadily increasing in the world. According to the data [1] approximately 200,000 tons of this type of products (approximately 450 million pounds/year) were manufactured and delivered in sale during 2008.

The expansion of bar processing of food products is related to the fact that using the method of cold preservation the prevention of microbiological damage is possible. It has been proved during the repeated researches that the barometric effect of the pressure of 600 MPa at 20°C during 180 sec is capable to liquidate causative agents of listeriosis (*Listeria monocytogenes*) in meat and meat products, and also to inactivate other life-threatening microorganisms - colibacillus (*E. coli*), salmonellas (*Salmonella*), cholera vibrio (*Vibrio*), the most of species of mold mushrooms and pathogenic bacteria [3]. Today the considered technology is applicable only for inhibition of processes of growth and reproduction of vegetative forms of bacteria, however the combination of pressure and temperature is capable to provide the inactivation of spores of microorganisms as well. Thus, for example, the spores of *Clostridium botulinum* and some representatives of the sorts *Bacillus* and *Clostridia* can be destroyed as a result of synergic effect of the temperature and barometric factor. Such an effect provides to reduce a thermal effect by means of the pressure additionally imparted to the system [4–6]. The resistance of sporous forms is much higher than vegetative ones because of the presence of a serious protective mechanism in the first ones. Thus, it is known it is necessary to put a product under the pressure of 300–400 MPa at 25°C for several minutes to deactivate yeast, however, to destroy yeast ascospores a higher pressure and a longer effect is required. Spores of *Clostridium botulinum* are considered the steadiest among bacterial pathogenic spores, and *Bacillus amyloliquefaciens* disputes – among nonpathogenic ones [5].

It has also been proved that the pressure over 200 MPa in the temperature condition not above 45°C is capable to inactivate effectively the vegetative forms of practically all pathogenic microorganisms and those which spoil the food with no effect on flavor characteristics [7]. However, it is important to note that the efficiency of process depends, to a greater degree, on the type and complexity of the organization of microorganisms, the chemical composition and pH of the processed environment, and also on water activity.

Gram-negative bacteria are more sensitive to the effect of high pressure, than gram-positive ones are. The barometric effect causes the destruction of cellular membranes and intracellular proteins playing the major role in the activity of microorganisms, this all provides the degradation of cellular structures and final destruction of a cage in general. The shift toward the acidic pH and the increase in pressure have a synergy effect during the elimination of microorganisms. Due to the increase in acidity of the environment, the inhibition of activity of water molecules occurs which provides a considerable delay of processes of inactivation induced by extra-high pressure [4]. Today the technology of high hydrostatic pressure includes two main methods – a batch and a semicontinuous one. Of practical use is, mainly, the batch technology that presumes that the packed lot of goods is placed into the chamber, then is hermetized and filled with a transfer medium (water or other low-molecular liquids). The targeted pressure imparted to the environment is transferred to the elastic walls of packing and thereof the compression of product [8, 9] occurs. The semicontinuous methods are not at all perfect today, both in the power and economic aspects. They have been created for the purpose of implementation of direct compression of liquid foodstuffs.

Fig. 1 presents the ratio between various classes of foodstuffs (%) the technology of high pressures can be applied to today.

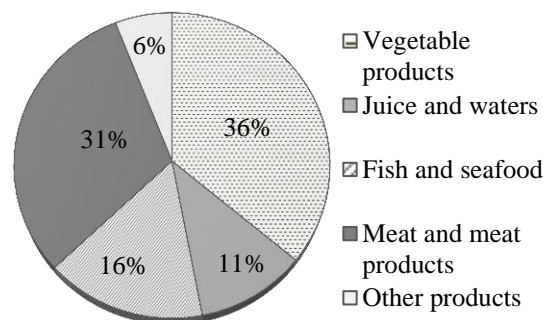


Fig. 1. Industrial use of high hydrostatic pressure for processing of different groups of foodstuffs [10].

It is established that during the processing under the pressure of 130 MPa there is inhibition of growth of microorganisms in beef for a week, the color of meat improves, the gained effect remains for 3 days at a storage temperature +4°C [9].

As a result of the researches, Han J.M. and Ledward D.A. found out that the rigidity of muscular tissue of beef increases with the increase in pressure from 200 to 800 MPa (at a constant temperature from 20 to 40°C), but considerably decreases during the use of pressure at a level of 200 MPa (at a temperature of 60 and 70°C) [10]. A Bai Y. and coauthors noted that after processing under high pressure (300–700 MPa) within 20 min. considerable changes of organoleptic properties of meat can be observed. They also recorded some modifications in the microscopic structure of myofibrils of the muscular tissue of cattle and mutton [11]. Qin H. and others proved that the activity of

calpains in the course of bar processing decreases, but the activity of acid and alkaline phosphatases does not significantly differ from the values of control samples. The pressure of about 100–200 MPa is capable to inactivate calpastatin (an inhibitor of activity of calpain) more rapidly than calpain itself is [12].

L.G. Vinnikova obtained some quite interesting results. She chose the processing under the pressure in the interval from 500 to 700 MPa during 30–60 sec as an optimum technological mode, which provided to inhibit the activity of acid phosphatases when imparting the maximum pressure (700 MPa) to the samples and thus to reach full culinary readiness with the organoleptic indicators corresponding to a boiled meat product. The mass losses of the target product in the course of processing were also reduced by 35% in comparison with the thermal effect. Thus, almost a hundred percent outcome of the finished product was stated [13].

As for the cost of modern equipment for processing, it varies from 500 000 to 2.5 million dollars depending on the power and extent of automation. The internal volume of a vessel varies from 30 to more than 600 liters [14].

The use of high hydrostatic pressure in food industry becomes more and more demanded every year. The interest of use of this particular technology is that it is capable to inactivate the action of microorganisms and fermental complexes without the inhibition of the energy and biological value.

Today one of the priority directions of state policy in the field of healthy nutrition and replacement of foreign food technologies is the development and integration of the foodstuffs enriched with irreplaceable micronutrients and also of new domestic processing equipment for food industry into production. The provision of safety of the biologically active agents put into the formulation throughout the entire period of storage deserves special attention.

The aim of work is the research of effect of bar processing on the periods of storage of boiled sausages enriched with vitamins, the assessment of efficiency of their use for correction of the state of health of students and the development of equipment alternative to import equipment capable to produce the pressure not less than 1200 MPa providing to use it in food industry.

OBJECTS AND METHODS OF STUDY

The objects of researches were boiled sausages in a nylon cover "Amilyuks" with a period of storage of 4 days at a temperature from 2 to 6°C, enriched with the vitamin premix 730/4 produced by ValetokProimpeks, CJSC in the amount of 150 g per 100 kg of the basic raw materials.

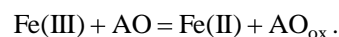
The control samples of boiled sausages enriched with vitamin premix were not processed under high pressure. The test samples of boiled sausages enriched with vitamins were processed after the end of the technological process under the pressure of 800 MPa within 3 minutes at a temperature of 0 ... +4°C by means of an experimental plant – a hydrostat (Fig. 2) with the following technical characteristics: the process pressure is 800–1000 MPa; the maximum pressure is 1200 MPa, the time of process stabilization is 2 to 3 min, the process liquid is a mix of industrial oil and glycerin.



Fig. 2. High pressure plant (hydrostat).

The safety of boiled sausages was estimated according to the organoleptic and microbiological indicators and pH, the content of vitamins in the product was researched in 8 and 16 days of storage.

The microbiological indicators are in accordance with GOST R 54354-2011 "Meat and meat products. General requirements and methods of microbiological testing", GOST 31747-2012 (ISO 4831:2006, ISO 4832:2006) "Food products. Methods for detection and quantity determination of coliforms". Using the pH-potentiometric method, vitamins – using the fluorimetric method. The antioxidant activity – using the potentiometric method. The shift of potential Pt electrode made using screen-printing technique in the mediator system $K_3[Fe(CN)_6]/K_4[Fe(CN)_6]$, observed during the administration of antioxidants (a sample) into the solution was the source of information of antioxidant activity. This shift occurs due to the change of ratio of the oxidized and reduced forms of components of mediator system as a result of the following reaction:



The researches were performed using a lab-scale plant in Institute of Metal Physics, Ural Department of the RAS (Yekaterinburg) and at the Department of Food Engineering of Ural State Economic University (Yekaterinburg).

The statistical processing of results was carried out with the use of the standard computer programs Microsoft Excel XP, Statistica 8.0.

RESULTS AND DISCUSSION

Table 1 presents the organoleptic indicators of boiled sausages in 8 and 16 days of refrigerating storage at a temperature of +4°C.

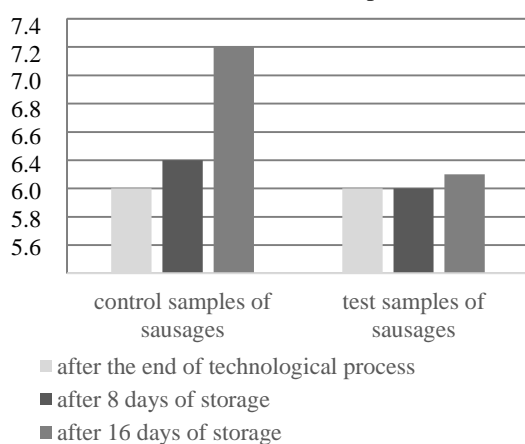
It follows from the data of Table 1 that after 8 days of storage the control and test samples of sausages conformed to the requirements of the Technical regulation of the Customs union "About safety of meat and meat products" (TR TS 034/2013). In 16 days of storage the control samples of sausages had a slippery and damp surface, lost their elasticity, differed in an ammoniac smell while the test samples conformed to the requirements of the regulating documentation.

Table 1. Organoleptic indicators of control and test samples of boiled sausages in 8 and 16 days of refrigerating storage at a temperature of +4 °C

Parameter	Group	
	Group 1 (control)	Group 2 (test)
After 8 days of storage		
Appearance	Bars with a clean and dry surface	Bars with a clean and dry surface
Consistence	Delicate and juicy	Delicate and juicy
Color and look in the cut	Pink, uniform mincemeat, evenly mixed	Pink, uniform mincemeat, evenly mixed
Smell and taste	Peculiar to this type of product, without foreign flavor and smell, with the aroma of spices, moderately salty	Peculiar to this type of product, without foreign flavor and smell, with the aroma of spices, moderately salty
After 16 days of storage		
Appearance	Bars with a clean and slippery surface	Bars with a clean and dry surface
Consistence	Delicate, juicy and less elastic	Delicate and juicy
Color and look in the cut	Pink, uniform mincemeat, evenly mixed	Pink, uniform mincemeat, evenly mixed
Smell and taste	Ammoniacal	Peculiar to this type of product, without foreign flavor and smell, with the aroma of spices, moderately salty

Researches of pH of sausages in the course of their storage (Fig. 3) have been performed.

Fig. 3 shows that the shift of pH to the alkaline side in the course of storage of control samples of boiled sausages is noted; the most significant changes are in the control samples which testifies an active activity of the residual microflora in a foodstuff providing proteolysis and, respectively, the accumulation of nitrogenous bases. The obtained data are coordinated with the results of organoleptic (Table 1) and microbiological researches. An authentic increase in pH has not been noted in the test samples.

**Fig. 3.** Dynamics of pH of boiled sausages during their storage.

A significant increase in the content of mesophilic aerobic and facultative and anaerobic microorganisms has been noted in the control samples of boiled sausages. The quantity of bacteria was 1.6×10^3 in 8 days of storage and 2.7×10^3 in 16 days which exceeds the requirements of TRTS 034/2013. The processing of boiled sausages under high pressure provided to receive a sterile foodstuff. Mesophilic aerobic and facultative and anaerobic microorganisms in the test samples of sausages have not been found during the entire period of storage.

Table 2 presents the content of B₁, B₂, PP and C vitamins in the course of their storage.

It follows from the data of Table 2 that in the course of storage of boiled sausages enriched with vitamin premix a decrease in the quantity of vitamins both in the control and test samples is noted, in particular, after 8 days of storage the quantity of B₁, B₂, PP and C vitamins decreased by 5.3%; 1.3%; 2.7% and 13.6% in the control samples. After 16 days of storage the content of PP and C vitamin in the control samples of sausages authentically decreased by 17.6% and 93.4% while in the test samples it decreased by 4.1% and 12%. It follows from the obtained data that the processing of boiled sausages, enriched with vitamin premix, under the high pressure of 800 MPa does not only destroy the vitamins of mincemeat and premix, but also provides to keep them safe for the entire period of storage.

Table 2. Content of B₁, B₂, PP and C vitamins in the course of production and storage of boiled sausages enriched with vitamin premix, mg/100g

Boiled sausages	Content of vitamins											
	After the end of technological process				After 8 days of storage				After 16 days of storage			
	B1	B2	PP	C	B1	B2	PP	C	B1	B2	PP	C
Control	0.57 ± 0.05	0.75 ± 0.08	7.4 ± 0.9	30.1 ± 0.7	0.54 ± 0.05	0.74 ± 0.07	7.2 ± 0.9	28.0 ± 1.1	0.53 ± 0.05	0.73 ± 0.05	6.1 ± 0.8	2.0 ± 0.9*
Test	0.58 ± 0.04	0.75 ± 0.07	7.4 ± 0.5	30.0 ± 0.7	0.56 ± 0.05	0.75 ± 0.08	7.2 ± 0.8	28.2 ± 1.0	0.55 ± 0.05	0.73 ± 0.07	7.1 ± 0.9	26.4 ± 1.0*

The microbiological decay of boiled sausages begins even prior to lipide oxidation, however the information of the resistance of boiled sausages to oxidation is absent in this case. In the course of oxidation of lipide components there is a rancidish taste, the color and consistence worsen and the nutrition value decreases. The process of autooxidation of lipids, during which unsaturated fatty acids react with oxygen with the formation of acylhydroperoxides or peroxides of fatty acids, proceeds according to a free radical mechanism. The substances that block or slow down the process of oxidation of lipids are called antioxidants. In this regard, we investigated the antioxidatic activity of boiled sausages (AO). As a result of researches, it has been established that the test samples of sausages had a higher AOA (0.42 ± 0.03 mol equiv/dm³), which is authentically 71.3% higher (** $P \leq 0.01$) than the AOA of the control samples (0.12 ± 0.04 mol equiv/dm³). The obtained data are explained by the presence of vitamins C and E, which have an antioxidatic action, in the vitamin premix 730/4 and by the high stability of vitamins in the test samples of boiled sausages.

The research of assessment of efficiency of use of enriched sausages in student nutrition was performed in South Ural State Agricultural University at the department of physical training and sport in the academic year 2015–2016. Two groups of students (young men) of the first course at the age of 18–19 were formed. The criterion for including in the research was the voluntary written consent to the participation in the experiment, the provision of necessary personal health information about themselves, the accommodation in the dorm, the meals in the university canteen and attending classes of physical culture. The criterion for dismissal was the incidence of acute infectious diseases at the time of the research or within 30 days prior to the experiment, the use of vitamins and mineral substances.

Table 3 presents the scheme of researches.

The assessment of nutritional level of students was performed according to the methodical recommendations (MR) of 2.3.1.2432-08 “Norms of physiological needs for energy and feedstuffs for various groups of the population of the Russian Federation”. The statistical processing of experimental data was performed by means of the computer program Statistica-6. The assessment of state of health of students was performed with the use of method of questioning and research of the antioxidant activity of blood and the indicators of a cellular link of immunity according to the standard techniques.

The questioning of students regarding the assessment of their food is performed according to a five-mark grading system. It is established that only 8% of the students estimate their food as excellent,

35% – as good, 45% – as adequate and 12% – as inadequate. It should be noted that 27% of respondents complained about the state of their health. They complained about frequent headaches, rapid fatigability, weakness, drowsiness, a decrease in working capacity, frequent acute respiratory diseases (ARD), stomach pains, the lability of arterial pressure and others. The obtained data are coordinated with the assessment of actual diet where there is lack of the vital micronutrients.

The criterion of the state of health of students are some indicators of antioxidant activity: antioxidant activity (AOA), the content of enzyme catalase (C) and protein ceruloplasmin (CP) in the blood of students.

An authentic increase in antioxidant activity by 26.7% in the blood of students of the test group is noted against a background of the use of vitaminized sausages. Similar changes are noted in the content of catalase and ceruloplasmin. Thus, the amount of catalase and ceruloplasmin increased by 19.2% and 16.1%. The authentic changes of indicators of antioxidant protection of the organism of students of the control group are not noted. An increase in working capacity and decrease in fatigue at a background of application of vitaminized sausages is noted in the questionnaires of 61% of students of the test group.

In view of the fact that the foreign equipment for processing under high pressure in the conditions of all-round compression is expensive, it is reasonable to manufacture a domestically produced hydrostat that would not yield to foreign analogs in technological parameters. Schematically a high pressure hydrostat is presented in Fig. 4. To manufacture the hydrostat strength calculations of the hydraulic cylinder of the compression chamber have been performed.

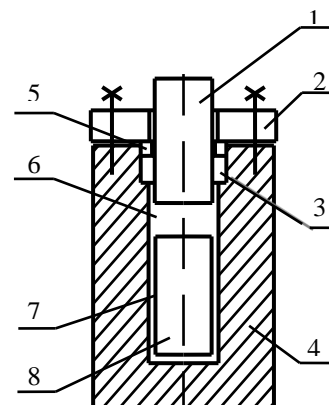


Fig. 4. Schematical design of a high pressure hydrostat for processing of objects with a liquid in the conditions of all-round compression: 1 – punch; 2 – flange; 3 – sealing; 4 – working chamber; 5 – washer; 6 – process liquid; 7 – cover; 8 – object of the research (product).

Table 3. Scheme of researches

Group	Number of students in the group, persons	Age of students, years	Correction of a diet by vitaminized boiled sausages	Dose and frequency of use of hematogen
Control	20	18–19	–	–
Test	20	18–19	Inclusion of vitaminized boiled sausages in the diet	100 g daily within 20 days

Strength calculations of the compression hydraulic cylinder of the high pressure hydrostat. Calculation of pressures in the compression hydraulic cylinder of the hydrostat.

Design pressure according to the condition of strength P_{des1} , MPa

$$P_{des1} = P_{1max} - P_{atm}, \quad (1)$$

Where P_{1max} is the maximum excessive pressure in the hydraulic cylinder of the hydrostat, MPa; P_{atm} is the atmospheric pressure, MPa.

For calculation we take $P_{1max} = 1200$ MPa; $P_{atm} = 0.1$ MPa.

Then the design pressure according to the condition of strength as per (1)

$$P_{des1} = 1200 - 0.1 = 1199.9 \approx 1200 \text{ MPa.}$$

As the pressures less than the atmospheric pressure are not supposed to be in the hydraulic cylinder of the hydrostat, the calculation for the chamber is performed only under the terms of strength.

Calculation of thickness of the wall of the hydraulic cylinder according to the strength theory. The cylinder is made of autofretted high-tensile steel of the brand "O-AB".

Design thickness of the wall of the hydraulic cylinder of the compression chamber S_1 , mm:

$$S_1 = S_{des1} + C_1, \quad (2)$$

where S_{des1} is a design thickness of the wall of the hydraulic cylinder, mm; C_1 is an addition to the design thickness considering the process of material corrosion, mm;

Addition to the design thickness considering the process of material corrosion C_1 , mm

$$C_1 = A \cdot t, \quad (3)$$

where A is the design rate of corrosion of the material of construction, mm/year; t is the planned service life of the hydrostat, year.

For calculation we take $t = 10$ years and $A = 0.005$ mm/year according to the standard for vessels and devices under pressure.

$$C_1 = 0.005 \cdot 10 = 0.05 \text{ mm/year.}$$

Design thickness of the wall of the hydraulic cylinder of the compression chamber S_{des1} , mm

$$S_{des1} = \max\{S_{n1}; S_{stab1}\}, \quad (4)$$

As the design thickness of the wall of the hydraulic cylinder the greatest of the received values under the terms of strength S_{n1} , mm, and stability S_{stab1} , mm, is chosen.

As the calculation under the terms of stability is not performed, then $S_{des1} = S_{n1}$.

Design radius of the hydraulic cylinder of the

compression chamber under the terms of strength R_c , cm

$$R_c = R_0 \sqrt{\frac{\sigma_{tens} + 0.4P_{stab}}{\sigma_{tens} - 1.3P_{stab}}}, \quad (5)$$

where R_0 is the internal radius of the body of the hydraulic cylinder sufficient for the placement of samples of foodstuffs and semifinished products in it. We take $R_0 = 100$ mm; σ_{tens} is the admissible tension of material of the body, for autofretted high-tensile steel "O-AB" $\sigma_{tens} \geq 50$ MPa; P_{stab} is the design pressure of process liquid ($P_{stab} = 1.2 P_{des}$).

$$P_{stab} = 1.2 \cdot 1200 = 1440 \text{ MPa,}$$

$$R_c = 10 \sqrt{\frac{5000 + 0.4 \cdot 1440}{5000 - 1.3 \cdot 1440}} = 13.35 = 14 \text{ cm.}$$

Design thickness of the wall of the hydraulic cylinder S_{n1} , cm

$$S_{n1} = R_c - R_0, \quad (6)$$

$$S_{n1} = 14 - 10 = 4 \text{ cm} = 40 \text{ mm.}$$

Design thickness of the wall of the hydraulic cylinder of the compression chamber, S_1 , mm

$$S_1 = 40 + 0.05 = 40.5 \text{ mm.}$$

Round the received value to the nearest standard value $S_1 = 42$ mm. The performed calculations provide to start designing a high pressure hydrostat for processing of products in the conditions of all-round compression.

As a result of the performed complex researches of indicators of freshness and safety of vitamins in boiled sausages it has been established that the samples processed with a high pressure of 800 MPa within 3 min. after 16 days of storage conformed to the requirements of the Technical regulation of the Customs union "About safety of food products" (TR TS 021/2011). The high pressure processing of the boiled sausages enriched with vitamin premix makes the foodstuff sterile as a result of a high pressure bactericidal effect on the microbic cells, prevents proteolysis, saves the vitamins, having an antioxidant effect which provides the weakening of processes of oxidation of lipidic components. The received results show that the use of high pressure in technology of storage of boiled sausages provides an increase in the periods of their storage. Against a background of daily use of boiled vitaminized sausages within 20 days the improvement of the state of health of students is noted, in particular, the antioxidant activity of blood and catalase enzyme authentically increases and the amount of ceruloplasmin protein increases. The obtained data are coordinated with the results of questioning. 61% of students note an increase in working capacity and improvement of the general condition of their organism.

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