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II.2. PHYTO-FEED ADDITIVES PRODUCTION: TECHNOLOGICAL ASPECTS AND BIOLOGICAL VALUE

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Abstract

The results of the qualitative assessment of the final products of the technology include: mowing of vegetative plants, such as alfalfa, amaranth, clover, pea-oat mixture; grinding; wet fractionation with obtaining marc and juice; thermal coagulation, filtration of juice, drying of the chloroplast fraction by spraying or in a vibro-boiling layer.In the process of transformation of vegetative plants, biologically active phyto-feed additives are obtained as the final product. During the production of which the processed phytomaterials were influenced by regime processing parameters related to temperature, processing of time on certain stages, acidity, etc. A computational method developed in the paper for determining the biological value of phyto-feed additives is implemented taking into account the importance of protein efficacy ratio.

Key words*: phyto-feed additives, protein quality, heat treatment, solubility, protein digestibility, process parameters*.

INTRODUCTION

The technological chain of protein feed production - processing of the initial mass-feed additive includes a number of complex interrelated processes that affect its final result (the quality of the feed produced, its quantity and energy consumption). The process of mechanical dehydration of plant mass (wet fractionation) can be considered as one of the main processes (Colas et al., 2013). Finally the process flow is divided into two kinds of the production - juice and pulp. The structural and functional analysis of the technology processes made it possible to draw preliminary conclusions. Each process of this complex technological system has input, output parameters and control actions. All these components are closely interrelated and determine the functioning of the process. The interconnection of processes in such a technological system is associated with a significant impact on the final result in case of serious violations in one of them. The technology for processing pomace includes several processes: drying by thermal methods or active ventilation, mixing with other feed components and briquetting of the mixture, production of haylage or silage feed for the laying of the pulp deposited using the methods of canning. Juice processing includes a number of complex processes that have a greater impact on the quality and quantity of the final product-biologically active feed additives. These processes include three stage: coagulation of proteins in juice plants (alfalfa, amaranth etc.), separation of the coagulate into a chloroplast paste-like fraction and serum, spray drying or in a vibro-boiling layer, depending on the production requirements. Classification of processes by their functional purpose, means of implementation, types of operations performed, nature, mode and level of regulation, give possibility to more accurately represent this complex technological structure of relationships. The intermediate type of feed material is the raw material for the next phase of the resource cycle. The end result of the entire technological system is the product. The processes associated with the heat effect on the processed product determine both the quantitative and qualitative side of the feed produced, related to the absorption of nutrients (supplements) by the animal and poultry body.

A large number of factors affect the quality of the final product in the technological complex of production of biologically active feed additives from green plants. The main factors are the time

spent on mowing raw materials before processing, the conditions for processing them in technological flows, the nature of impacts and the degree of oxidation by the working bodies of machines, the sanitary condition of the product, and the modes of heat treatment of feed raw materials (Kalli et al., 2018). The conditions of heat treatment of products have a significant impact on their quality (Pęksa & Miedziank, 2014). It depends not only on temperature and exposure time, but also on the residual content of phenols and carbohydrates in the material being processed (Seczyk et al., 2019; Rong et al., 2013). The leaf protein concentrate (LPC) can be separated from deproteinised juice (DPJ) by filtration through cotton cloth (Badar & Kulkarni, 2011). It was that the process of Green Crop Fractionation (GCF) can be employed for the preparation of feed grade pressed crop residue (PCR) and food grade leaf protein concentrate (Rathor, 2016). Toxic constituents like nitrates and oxalates, accumulated in the foliages of several plant species, were generally removed in the DPJ and as a result of which the protein concentrate (PC) and LPC contained safer levels of these toxic elements in view of their value as either feed or food (Sayeed & Gogle, 2002). In this way green foliage can be fractionated mechanically into three fractions: (i) fibrous pressed crop, (ii) leaf protein concentrate and (iii) deproteinised juice. Leaf concentrate is an extremely nutritious human food, containing approximately 50% (dry weight) high quality protein, together with numerous micronutrients, principally b-carotene, vitamins B_6, B_9, E and K, plus iron, calcium and magnesium The green juice is a raw material for high quality fodder proteins, cosmetic proteins, human nutrition or platform chemicals like lactic acid and lysine (Arlabosse et al., 2011). Pumpkin and amaranth leaves recorded the highest protein yield (10.5-11.75%). The protein contents in the leaves of sweet potato, cowpea, cabbage and sugar been were much lower (Ghaly & Alkoaik, 2010). The data of chemical analysis and calculation of the total amount of exchange energy in the feed, cannot fully give reliable information about its quality. Biologically active phyto-feed additive and by-products of technology might have high digestion by the animal as well as low one. This fact is the main qualitative criterion of obtaining feed, taking into account the influence of technological processing regimes onto feed quality indicators. It is necessary to choose such regimes in technological processes, which at the final stage of the whole chain would guarantee saving biologically active nutrients (amino acids, vitamins, minerals) and high digestibility of feed additives as well. The most important components in the feed are protein, fats, fiber, fate soluble vitamins, so the influence of technological regimes on the quality of feed can be assessed by the degree of protein digestion by animals. Biologically active feed additives are rich in carotene, vitamins, and protein. The amino acid composition is similar in composition to the amino acids of animal origin. Technological equipment and innovative solutions, their interconnections in processes, operating parameters of equipment for the production of biologically active feed additives and the organization of the raw material base, technical means for cleaning, transportation and processing into the main products, represent a promising direction in the industry of industrial feed production with effective use in poultry and animal husbandry.

MATERIALS AND METHODS

The methodology for obtaining biologically active phyto-feed additives is based on the fact that the plant cell protein is contained in all intracellular formations - the cytoplasm, chloroplast, mitochondria, and nucleus. The most valuable part of the intracellular protein is ribulose-1.5-biphosphate carboxylase.

Intracellular protein of green plants is a rich source of protein and biologically active additives. The essence of the technology consists in mowing and grinding plants, pressing green juice by pressing methods, coagulation for the formation of chloroplast and cytoplasmic flakes, separating them as a paste by filtration or centrifugation. Green mass extracts are a by-product of this technology. The main components of pomace are fiber and serum after filtration.

The second stage of fractionation provides the production of green mass of seeded herbs, biologically active additives in the form of juice, paste, dry concentrate. These products are full-fledged substitutes for animal proteins and soy

meal and are used as a dietary supplement in the feeding diets of poultry, piglets and calves.

The processing of green juice and the use of the press residue ensure the complete preservation of the biological crop. The stages of processing include fractionation of leaf-stem biomass, the production of paste drying and production of dry biologically active feed additives for poultry diets of various breeds and ages, young poultry, piglets, calves, sheep and goats, broilers and breeding poultry. The kinetics of protein-carbon-phenol complexes formation foresee that the effect of heating on processed products should be determined by a comprehensive study of a scope of biochemical indicators of products obtained under different conditions. One of the main factors determining the quality of the protein is its digestibility in the gastrointestinal tract. It is well known that the correlation between the amino acid composition of the protein and its biological value occurs only if there is a sufficient rate of proteins digestibility by digestive enzymes.

From the practice of using feed in animal husbandry, negative correlation is considered the level of digestibility of dry matter in the diet, animal and amount of its consumption, especially highly productive ones. For example, dairy cattle need to be feed by fodders with a digestibility of dry matter at least 65%. In addition, the main indicator of the feed diets usefulness is the balance in essential nutrients. Hence, there are high demands to industrial technologies, equipment and management in feed production, in order to obtain feed with high exchangeable energy. The amount of metabolizable energy in the feed can be determined by the following regression equations: for pigs:

ME = 20.85 dp + 36.63 df + 14.27 dfb + 16.95 dnfe; for fowl:

ME = 17.84 dp + 39.78 df + 17.71 dfb + 17.71 dnfe; for cattle:

ME = 17.6 dp + 31.23 df + 13.63 dfb + 14.78 dnfe; where: dp - digestible protein, g;

df - digestible fat, g; dfb - digestible fiber, g;

dnfe - digestible nitrogen-free extractives, g.

The total biological value of the concentrate of phyto-feed can be determined by a calculation method based on the value of the protein efficiency ratio (PER), determined from the change in body weight of experimental animals (Bhilave et al., 2012). PER takes into account the ratio of weight gain by animals to the amount of edible protein when feeding standardized animals with standardized rations. The calculated method for determining the PER index considers the content of essential amino acids in a protein and its digestibility *in vitro*. Computed PER (C-PER), takes into account the digestibility of protein and casein, the content of essential amino acids and the score of each essential amino acid in the studied protein and casein. C-PER is calculated using the equations:

$$z = (SPC) \cdot 2.94 \cdot \frac{2.5}{2.94} \quad (1)$$

$$C - PER = -2.1074 + 2.8525 \cdot z - 0.4030 z^2$$

A qualimetry method is quite useful (Kuznetsov et al., 2019). It allows determining the quality characteristics of products based on the assessment of each one. Also it takes into account the weight ratio of the properties of the product. The mathematical model proposed of the qualimetric method is following:

$$K = M_a \sum_{i=1}^{i=l} m_{ai} K_{ai} + M_в \sum_{i=l+1}^{i=p} m_{вi} K_{вi} + M_c \sum_{i=p+1}^{i=q} m_{ci} K_{ci} + Md \sum_{i=q+1}^{i=n} m_{di} K_{di}, \quad (2)$$

where: n - features, characterizing the quality of products; M_a, $M_в$, M_c, M_d - the relative weight of each group of properties characterizing quality, $M_a + M_в + M_c + M_d = 1$ m_{ai}, $m_{вi}$, m_{ci}, m_{di} - the relative weight of each i-th property for each group of properties.

$$\sum_{i=1}^{l} m_{ai} = 1; \sum_{i=l+1}^{p} m_{вi} = 1; \sum_{i=p+1}^{q} m_{ci} = 1; \sum_{i=q+1}^{n} m_{di} = 1 \quad (3)$$

RESULTS AND DISCUSSIONS

The study of processes on stationary hydraulic presses in perforated cylinders, on auger mechanical presses was part of the task of experimental research on the fractionation of the green mass of alfalfa into juice and pomace. Also, the analysis of the process of juice separation under different modes of influence on the mass and finding dependencies that characterize this process was carried out. Program of studies provided that: the determination of the yield of green mass, the impact of interest remove moisture depending on the pressure, exposure time, initial moisture

content, volume of the processed mixture, removing plots of socket data a certain amount of weight depending on the application, stable load repeated impact: determine the impact of contact area and the magnitude, time pressure squeezing on the magnitude and intensity of the impact of the juice.

The results of the experiments showed that there is a close relationship between the initial humidity of the green mass of alfalfa and amaranth and the effectiveness of the dehydration process. As the mass humidity increases, the pressing capacity for juice and pulp increases, and the energy intensity decreases on the contrary. It is characteristic that a sharp increase in energy intensity is observed when a mass with humidity below 70% is dehydrated. Weak protein coagulation, the formation of small flakes (10-20 microns) is noted already at the acidity of the juice pH 5.6. Acidification of the solution to a pH of 4.05 contributes to more active protein deposition, the formation of a clear precipitate of coagulated chloroplasts. Increasing the heating temperature reduces the time required for protein coagulation. As a starting material, alfalfa juice was used with an initial temperature of 20 ... 25°C, a dry matter content of 11% and 3.2% protein. The steam pressure varied within $1.5 \cdot 10^5 - 6 \cdot 10^5 Pa$. The temperature of the heating steam (direct contact with the juice) was 140 ... 155°C, the steam consumption within 0.17 ... 0.23 kg/kg. The temperature of the coagulated juice varied within 45 … 90°C.The obtained data showed that for complete coagulation of proteins in the juice, it is necessary to extract the already coagulated juice for 50 ... 90 seconds. During this time, proteins are aggregated into larger a structural compound, which positively affects the separation by any means - centrifugation, filtration, and sedimentation. The analysis of the obtained experimental data showed that the operating modes of the centrifuge that satisfy the qualitative separation of the suspension into paste and brown juice are: - by the separation factor in the range of 1250 ... 1360; - by the second feed of the suspension -0.32 ... 0.35 l/s. Positive results on separation efficiency and changes in precipitation humidity are not achieved even when the feed is reduced to 0.12 l/s (for separation factors F_r = 420 and 698). When the separation factor increases to 903 ... 1360, the separation quality increases dramatically, the humidity of the chloroplast paste decreases, and the separation efficiency increases to 88.5%.Experimental studies of drying biologically active phyto-feed additives in a vibro-fluidized layer were conducted in a laboratory setting, simulating the camera vibro-dryers and to determine: the frequency and amplitude of vibration; the speed and temperature of the coolant; the drying time, the thickness of the dried layer. Chloroplast paste with a moisture content of 62 ... 36% was granulated through spinners with a diameter of 3 mm. The speed of the coolant varied within 1.4 ... 2.5 m/s, the amplitude of vibrations 5 ... 10 mm, the frequency of vibrations of the working cup 400 ... 700 fluctuations/min, the thickness of the granule layer varied within 20 ... 120 mm, the temperature of the heat carrier from 50 to 140°C. The results showed that with increasing time of heat treatment above 80°C and a process time more than 30 minutes - loss of carotene increase by 50%, and with increasing treatment temperature the losses increase even with a decrease in drying time. A short-term increase in the coolant temperature to 140°C at the initial moment of drying provides a sharp decrease in moisture to 420 g/kg, and in the future the drying intensity decreases. Determined time point, at which begins the growth temperature of the dried granules, the ingredients for a coolant temperature of 90 ... 140°C - 6 ... 9 min. Main time of removing moisture from the pellets is 20 ... 21 min, further - final drying. The modes that do not overheat the granules are determined, and the loss of carotene, protein and vitamins in the product is minimal. The found product characteristics make it possible to carry out a predicted energy calculation of heat for heating the material and time ranges of drying at which irreversible denaturation of protein does not occur. Various effects of coolant temperatures, both in magnitude and duration, allowed us to determine the nature of fermentation of leaf protein proteins with pepsin and trypsin, which can be used to judge the assimilation of the obtained products by the animal body. The qualimetry method is associated with experimental ones for estimating the values of M and m, which is associated with a considerable degree of subjectivity. It should be

emphasized that the considered methods, including the C-PER index, do not take into account some of the important features of the kinetics of protein hydrolysis by enzymes, as well as the solubility of biologically active phyto-feed additives. The total calculated indicator of the protein feeds quality should be described by the equation:

$$Q_{pgc} = \left(m_1 \frac{C - PER_{sample}}{C - PER_{standard}} + m_2 \frac{1}{n} \sum_{i=1}^{n} \frac{V^{sample}}{V_n^{standard}} \right) \varphi, \quad (4)$$

In equation (4), the parameter V takes into account the vitamin amount in the feed:

$$V = \frac{1}{n} \sum_{i=1}^{n} \frac{n_{sample}}{n_{standard}}, \quad (5)$$

where $n_{sample.}$ and $n_{standard}$ - the amount of vitamins in the sample and in the standard, respectively;

φ - factor taking into account the sanitary and hygienic indicators of the feed;

$$\varphi = \prod_{i-1}^{z} \varphi_i,$$

where $\varphi_i =$ { 0 at concentration of PC* ≤ δ ; 1 at concentration of PC ≥ δ }

z - number of accepted safety criteria;

*PC - permissible concentration;

$C\text{-}PER_{sample}$, $C\text{-}PER_{standard}$ - calculated indicator that takes into account the digestibility of protein in the feed, the content of essential amino acids, respectively, in the sample and standard;

$V_n^{sample}, V_n^{standard}$ - vitamin content in the sample and standard;

m_1, m_2 - coefficients of significance.

The hydrolysis of protein in the body of an animal can be described by the following equation:

$$p = a + b\left(1 - exp^{-ct}\right), \quad (6)$$

where ρ - the amount of hydrolytic material in a certain period of time (*t*);

a, *b*, *c* - constant in exponential equation.

Constant a - represents the rapidly digestible fraction; *c* - rate constant of hydrolysis fraction per 1 hour; *b* - the amount of material hydrolyzed in a specific period of time.

Since the criterion for evaluating the production modes of all processes of the technological system is the quality of the product produced, it is necessary to know the dynamics of the decomposition of dietary supplements, which must be obtained experimentally. In this case, the dynamics of the hydrolysis rate of the phyto-feed additives makes it possible to evaluate the effect of temperature regimes on the chloroplast proteins for dietary supplements. At the same time, the rate of digestion, as already noted, is an important indicator that determines the possible quota to substitute animal-derived proteins for plant ones in animal diets. Taking into account the determination of the availability of amino acids (N_i) to be attacked by enzymes, such as pepsin and trypsin, the amount of digested protein $S^{0}{}_{t_k}$ during time (t_k) can be described by the following formula:

$$S^{\circ}_{(t_k)} = \min_{1 \le n \le 8} \left(\frac{A_n}{F_n} \right) K_1 \cdot K_2 \sum_{i=1}^{20} \frac{A_i N_i}{100} \int_0^{\pi} \left[1 - exp\left\{ -\alpha_i (\tau - \tau_i) \right\} \right] \cdot d\tau, \quad (7)$$

Where: K_1 - protein content in a biologically active phytonutrient additive;

K_2 - protein digestibility;

A_n - essential amino acids content, in ideal protein (FAO);

A_i - amino acids content;

α_i - characteristic time;

τ - time;

t_i - the time of appearance of the amino acid in the hydrolyzate.

But this equation does not take into account the vitamin content of the feed and the kinetics of protein solubility, depending on the processing temperature. These parameters are included into generalized quality indicator K. The value of K will vary depending on the dynamics of the breakdown, changes in the amino-acids composition. All this is able to characterize the regimes under which the biologically active phyto-feed additive is produced. In this regard, taking into account the vitamin content in the product and the influence of the temperature parameters on the final product, the quality of the produced biologically active additive, should be described by the system of equations:

$$\begin{cases} K = \left[m_1 S(T,\tau) + m_2 V(T,r) \right] \varphi \\ S(T,\tau) = S^{\circ} exp\left(-\lambda_1(T)\tau \right) \\ V(T,\tau) = V^{\circ} exp\left(-\lambda_2(T)\tau \right) \end{cases} \quad (8)$$

where:
m_1, m_2 - the relative weight of the amino acid and vitamin portions in the feed;
$S°(t_к)$ - the amount of digested protein determined by equation (7);
T - temperature;
T - time;
V^o, $V(\tau)$ - the content of vitamins in the original feed and by the time τ after the start of drying, mg per kg;
$\lambda_1(T)$, $\lambda_2(T)$ - constants characterizing the breakdown of protein and vitamins at certain temperature.
The temperature dependence of the decay constant λ_1 (T) and λ_2 (T) in the simplest case is determined by the Arrhenius law:

$$\lambda_1(T) = \lambda_1^{\circ} exp\left(-\frac{E}{RT}\right)$$
$$\lambda_2(T) = \lambda_2^{\circ} exp\left(-\frac{E}{RT}\right) \quad (9)$$

where: E - activation energy of 600 J/mol;
R - universal gas constant equal to 8.314 J/(K mol);
T - temperature in °K.
The rate of isothermal denaturation can be described by the equation:

$$\frac{dx}{d\tau} = -K''(1-x) \quad (10)$$

where: x - degree of denaturation of the feed, calculated by the change in solubility;
K″ - denaturation rate constant.

CONCLUSIONS

The proposed method for assessing the quality of produced bioactive phyto-feed additives from green plants allows predicting the impact of thermal effects (heating the raw mass before processing, modes of coagulation, filtration and drying of additives, etc.) on changes in protein vitamins, biologically active components, content, in the final product. It could be used to predict the digestibility of phyto-feed additives by animals.

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