

# Sustainable valorisation of technical egg albumen: modelling and optimization of the coagulation process

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**Abstract.** Industrial egg processing assures more convenience and safety for the consumers but generates a significant quantity of by-products. The egg white adherent to the shell, referred as technical albumen (TA) is a liquid by-product containing valuable nutrients. The study investigates the influence of pressing force (PF), pH and dry matter concentration (DC) on the acid-thermal coagulation of TA by D-optimal design. The highest pressing efficiency (PE) was at 315 g/cm<sup>2</sup> P, 5.0 pH and 22% Dry matter (DM), on other hand 18.5% DM had no significant effect. In the same time the highest yield was at 5.0 pH, 351 g/cm<sup>2</sup> P highest and high 22% DM as well as, at 18.5% DM and 351 g/cm<sup>2</sup> P and 4.8 pH. The performed D-optimal central composite design (CCD) combined with response surface methodology (RSM) and mathematical optimization established the optimal conditions of the process at 22% DM, 4.80 pH and 333.3954 g/cm<sup>2</sup> P.

## 1 Introduction

Eggs are a one of the most high-quality nutrition foods in human diet [1]. They are defined as the cheapest animal source of protein [2]. Beside their nutritional value, eggs are used as foaming, emulsifying agents, as well as flavouring additive in variety of food products [3]. The industrial processing of eggs (e.g. dried, liquid or frozen egg products) offers economic benefits by extending the shelf life of the product and facilitating its transportation and storage [4]. The applied heat treatment reduces microbial contamination and destroys pathogens [5]. Approximately 30% of the consumed eggs in developed countries are processed egg products. Meanwhile those rates are reaching up to 50% in some European Union countries [1,6].

The industrial egg processing generates a ton of by-products daily. According to the Environmental Protection Agency, egg processing is ranked as the 15th major food industry pollution problem [7]. Egg by-products mainly include eggshells and membranes, wastewater from the egg processing, liquid waste products from egg breaking facilities, and eggs inadequate for human consumption [8]. All animal by-products (ABPs) pose a risk to humans, animals and the environment. For this reason, European legislation sets strict requirements about handling, and decontamination before further use. However, those by-products are rich in protein, providing all essential amino acids, fats - as source of energy as well as vitamins and minerals, mainly phosphorus and calcium and B vitamins [9]. In the context of circular economy, industry is expected to work in a closed loops of resources and follow the 3 R principle – reduce, reuse and recycle [10]. The best solution for these by-products is their reuse as raw

materials for new products, benefiting not only the economy and also the environment [11].

The second largest by-product is the egg white adhering to the shell, also known as technical albumen. The dry matter content is mainly proteins, which have been found by SDS-PAGE to be similar to the proteins in egg white [12]. The water content is about 80%, the proteins are 12%, with all amino acids found, the fats are 5-8%. The chemical composition of the industrially produced technical egg white varies depending on the speed of the egg breaking machine, the temperature, and the viscosity of the egg white [13]. The most convenient way to increase the shelf life of liquid egg products are to dry them into a stable dry powder form using the spray dryers. Due to low moisture content and low water activity obtained, powder is stable and resistant to microbiological spoilage and oxidative changes. This technology has already been involved into egg by-products processing, mostly in the production of animal feed. The cons of this technology are high investment costs of the equipment, high operating costs due to the requirements for drying air temperatures above 170°C [14-15].

The most crucial factor affecting drying is the initial moisture content of the raw material. Usually the final target moisture content of the product is below 10%. If the initial moisture content of the raw material is high, more moisture will need to be removed, which will cause a large heat impact on the product, increase processing time and fuel costs [16].

Previous studies describe a novel technology for TA processing, including a preliminary moisture reduction before drying. The method is based on the changed protein properties near the isoelectric point (*pI*), in other words the loss of electrical charge, causing aggregation

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and spontaneous release of moisture. Authors reported a significant difference upon using different coagulation agents over a wide pH range. It was found that the most suitable acid for processing TA, using the acid-thermal coagulation method is citric acid (CA). The expected highest yield, and the most suitable pressing method for processing TA, should occur at a pH range of 4.8-5.2 [13,17,18].

Therefore, the aim of this study is to model and optimize the coagulation process by evaluation of the influence of pH, dry matter of TA and pressing force on the yield and pressing efficiency on the processed TA by the acid-thermal coagulation method.

## 2 Materials and methods

### 2.1 Materials

The technical egg white/albumen (TA) was supplied by a registered manufacturer of pasteurized egg products (OVO-BUL Ltd., Pleven, Bulgaria). TA was obtained from the shells of broken eggs, during the regular production of egg products for human consumption, after centrifugation in a SANOVO SEC 360 centrifuge.

The initial dry matter of the TA was 22%. The remaining samples with dry matter of 15% and 18.5% were modelled by adding tap water to the achieve the desired dry matter concentration.

A 50% (v/v) solution of food grade citric acid was used for a pH correction.

### 2.2 Experimental design

The experiment was performed following the structured of a D-optimal central composite design (CCD) for three factors at two levels (Table 1).

The TA was processed according to the method of Saraliev et al. [18] with modifications. The pressing was executed in a purpose-built intermittent screw press (Figure 1). The size of the screen holes was 1 mm the diameter of the basket is 33 cm. Area of circle is 855 cm<sup>2</sup>. The coagulate was pressed with a compressive force of 200 kg, 300 kg and 400 kg, corresponding to a pressure of 234 g/cm<sup>2</sup>, 351 g/cm<sup>2</sup> and 468 g/cm<sup>2</sup>, respectively.

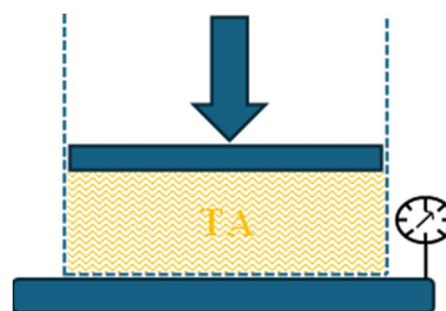
The pressing force was measured using a scale brand A Lift model T15 manufactured in PRC.

The pressing time of each sample was 7 minutes from the first serum release.

The pressed product was manually fragmented into particles up to 10 mm. The crushed coagulate was placed in 40/60 cm baking trays and dried in hot rack oven for 8 hours at a temperature of 80°C. The dry products were placed in plastic packaging and weighed on an electronic scale VEDIA VDS 15/30, manufacturer: Vedicom Ltd., Bulgaria.

**Table 1.** D-optimal central composite design (CCD)

Design point	x <sub>1</sub>	x <sub>2</sub>	x <sub>3</sub>	DM, %	pH	P, g/cm <sup>2</sup>
	Coded form			Natural form		
1	-1	-1	-1	15.0	4.8	234.0
2	1	-1	-1	22.0	4.8	234.0
3	-1	1	-1	15.0	5.2	234.0
4	1	1	-1	22.0	5.2	234.0
5	-1	-1	1	15.0	4.8	468.0
6	1	-1	1	22.0	4.8	468.0
7	-1	1	1	15.0	5.2	468.0
8	1	1	1	22.0	5.2	468.0
9	-1	0	0	15.0	5.0	351.0
10	1	0	0	22.0	5.0	351.0
11	0	-1	0	18.5	4.8	351.0
12	0	1	0	18.5	5.2	351.0
13	0	0	-1	18.5	5.0	234.0
14	0	0	1	18.5	5.0	468.0
15	0	0	0	18.5	5.0	351.0
15'	0	0	0	18.5	5.0	351.0
15''	0	0	0	18.5	5.0	351.0



**Fig 1.** Scheme of the pressing process

### 2.3 Production Parameters

The Pressing Efficiency (PE%)

The PE describes the product's ability to release liquid by applying pressure to it. It represents the ratio of the amount of liquid released during pressing to the initial amount of product expressed as a percentage. A larger value of EP indicates a lower amount of water in the coagulant and a better efficiency of the pressing process.

$$PE = \frac{\text{initial mass of the sample, g} - \text{pressed material, g}}{\text{initial mass of the sample, g}} \cdot 100, \% \quad (1)$$

### 2.4 Yield

TA is a raw material that has a different amount of dry matter. The yield depends on the initial amount of dry matter in the raw product, therefore the the yield is presented as a percentage of the theoretical maximum.

$$Yield = \frac{\text{final mass of the sample, g}}{\text{Initial mass of the sample, g} \cdot \frac{DM_{\text{raw}}}{100}} \cdot 100, \% \quad (2)$$

DM<sub>raw</sub> – Dry matter of raw product, %

## 2.5 Dry matter content

The dry matter content was determined by drying the samples in a moisture analyser Radwag MA.50, Poland at 104 - 105°C until constant mass was reached (ISO 1442: 1997).

## 2.6 Determination of pH value

For this purpose, a portable pH meter Hanna, HI99163 (Hanna Instruments, USA) was used, pre-calibrated with certified buffer solutions of 4.04 and 6.86.

## 2.7 Protein concentration

The protein concentration of the solutions was determined by the method of Lowry et al., [19] modified by Saraliev et al. [13].

## 2.8 Protein Separation and Identification

Aliquots diluted to 3 mg/ml protein content were mixed with a Laemlli Sample Buffer (Sigma-Aldrich, Co., St. Louis, MI, USA) in a 3:1 ratio, boiled, cooled, centrifuged, and 20 µl were injected into 10% polyacrylamide gels [20]. Protein separation was conducted at a constant voltage (200 V) and current (70 mA) using an Omni PAGA Electrophoresis System (Cleaver Scientific Ltd., Rugby, UK) [21]. The Precision Plus Protein Standards (Bio-Rad Laboratories, Inc., Hercules, CA, USA) was used as a protein marker. SDS-PAGE gels were processed with GelAnalyzer 23.1.1 software (available at www.gelanalyzer.com by Istvan Lazar Jr., PhD and Istvan Lazar Sr., PhD, CSc).

## 2.9 Statistical analysis

To evaluate the significance of independent variables (factors of the experiment) data was analysed using PROC GLM procedure in SAS version 9.4 (SAS Institute Inc., Cary, NC, USA). Least Square Means were compared using Tukey–Kramer post hoc test, and differences were considered statistically significant at  $p \leq 0.05$ . As a subsequent action was performed a response surface methodology (RSM) using JMP® Pro software version 17.0.0 (JMP Statistical Discovery LLC, Cary, North Carolina, U.S.)

The desirability function was used to optimize the process parameters according prior set target functions: maximum of both, dry matter content after pressing (DMC) as well as Yield.

## 3 Results and discussion

### 3.1 Raw material characterization

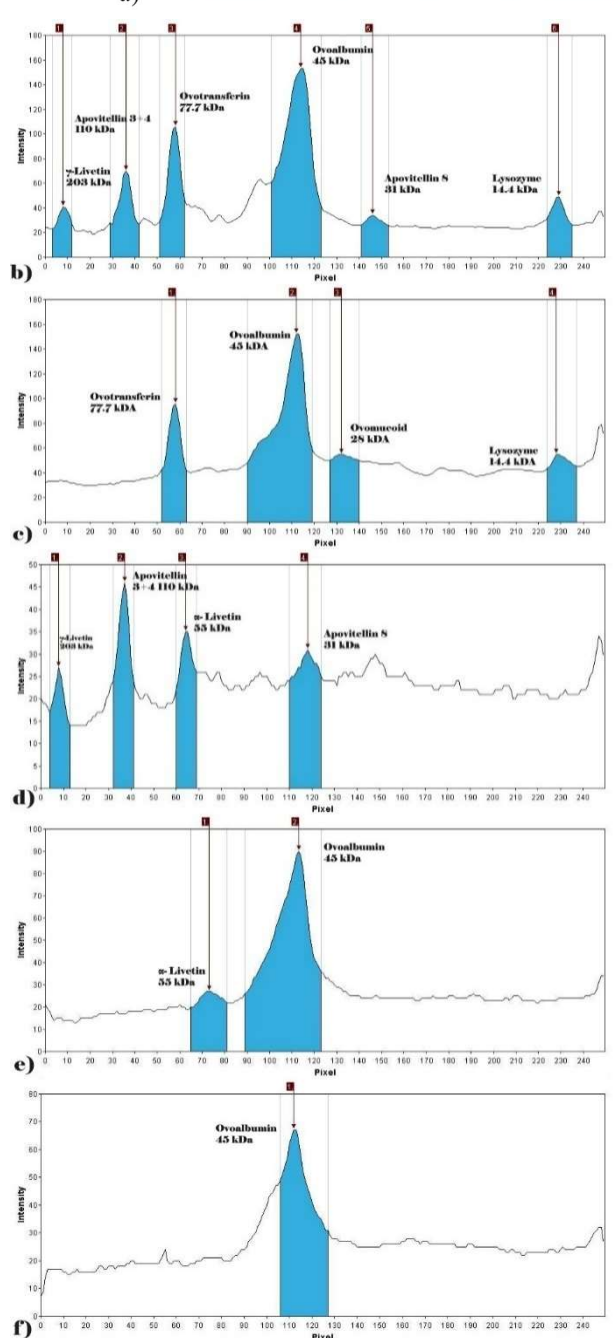
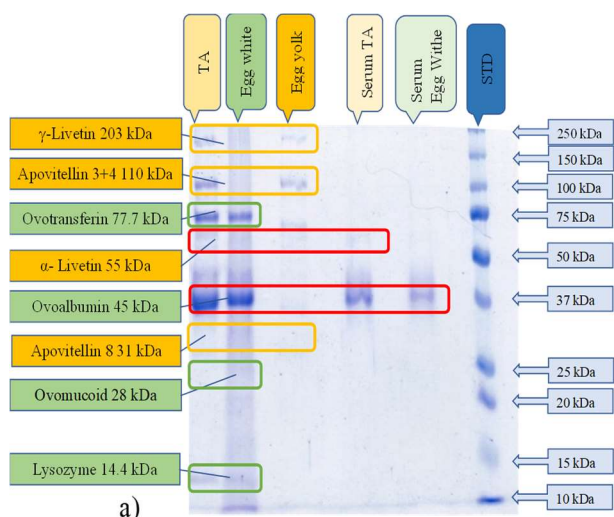
TA contains mainly proteins, which are the basis of the method of processing by acid-thermal coagulation. The protein profile of the raw material is important for the process and affects the production indicators PE and yield. The Figure 2 presents a SDS PAGE gel of TA as

well as serum released from the coagulate after pressing. A four of the major proteins in egg albumen. Ovalbumin (45 kDa,  $pI$  4.5, 54%), Ovotransferrin (76 kDa,  $pI$  6.1, 12%), Ovomuroid (28 kDa,  $pI$  4.1, 11%), Lysozyme (14.3 kDa,  $pI$  10.7, 3.4%) were found [12, 22]. Our findings confirm the reported presence of proteins with molecular weight of 76 and 45 kDa in fresh eggs from China [23]. Ji et al. [24] presented a protocol for the sequential extraction of egg proteins via pH adjustment, resin adsorption, ultrafiltration, and salt/ethanol precipitation. They report a purity of over 90% for lysozyme, ovalbumin, ovotransferrin, ovomuroid as well as over 75% for ovomucin. The same study reports an over 90% yield of lysozyme, over 80% of ovotransferrin and over 70% of the other proteins.

The proteins found in the egg yolk gel are plasma proteins.  $\gamma$ -Livetin 203 kDa and  $\alpha$ -Livetin 55 kDa are from the Livetin group of water-soluble glycoproteins. Apovitellin 3+4 110 kDa and Apovitellin 8 31 kDa are High-Density Lipoprotein (HDL) with antioxidant properties (Figure 2 d) [25-26].

Proteins from both egg white and egg yolk were detected in TA (Figure 2 a). Our result confirm the reported of presence of egg white proteins - Ovotransferrin (77.7 kDa), Ovoglobulin (49 kDa), Ovalbumin (45 kDa) and Lysozyme (14.3 kDa) as well as yolk proteins – $\gamma$ -livetin (203 kDa), Apovitellin 3 + 4 (110 kDa), Apovitellin-Vb (93 kDa), Apovitellin V (85 kDa), Apovitellin IV (68 kDa),  $\alpha$ - livetin (55 kDa ) and Apovitellin 8 (31 kDa) in previous study [13]. The proteins with the highest intensity in the TA diagram in Figure 2 b) are the egg white proteins Ovalbumin and Ovotransferrin. The processing method is based on thermal coagulation at a temperature above 85°C at the isoelectric point of the proteins, where the net electric charge is close to 0 and the repulsive forces are weakest. The  $pI$  of the predominant protein Ovalbumin is 4.5, and that of Ovotransferrin is 6.1, therefore the expected  $pI$  is slightly above 4.5 [22]. For this reason, we perform coagulation in the pH range 4.8-5.2.

The purpose of coagulate pressing is to separate the liquid fraction - serum, which contains mostly water as fraction of native proteins that were not precipitated (Figure 2e). The purpose of proper coagulation and pressing is to have as few dissolved substances in the serum as possible. The analysis of serum by TA (Figure 2 f) shows the presence of two proteins - Ovalbumin and with a very low intensity  $\alpha$ -Livetin. The intensity of the ovalbumin peak in the serum diagram from TA is lower than that in TA, indicating a decrease in serum concentration. The coagulation temperature of ovalbumin is the highest of all egg proteins – up to 77.5°C [27]. The processing temperature borderline was 85°C, leading to a full protein denaturation. On the other hand, a presence of ovalbumin in the serum could be due to its partial denaturation.



**Fig. 2.** SDS-PAGE gel and electropherograms: a) SDS-PAGE gel; b) Technical albumen (TA); c) Egg white; d) Egg yolk; e) Serum from TA; f) Serum from egg white

**Table 2.** PE, DMC and Yield values of TA processed by the acid-thermal coagulation method

DP	PE, %	DMC, %	Yield, %
1	24.83 <sup>bcd</sup>	27.52 <sup>cd</sup>	93.66 <sup>ab</sup>
2	20.25 <sup>defg</sup>	26.80 <sup>d</sup>	96.76 <sup>a</sup>
3	17.59 <sup>fg</sup>	24.48 <sup>h</sup>	91.32 <sup>b</sup>
4	16.38 <sup>gh</sup>	25.41 <sup>fg</sup>	96.20 <sup>ab</sup>
5	27.93 <sup>ab</sup>	28.95 <sup>b</sup>	94.44 <sup>ab</sup>
6	21.75 <sup>cdef</sup>	27.32 <sup>cd</sup>	96.76 <sup>a</sup>
7	26.21 <sup>abc</sup>	28.04 <sup>bc</sup>	93.66 <sup>ab</sup>
8	18.50 <sup>efg</sup>	25.61 <sup>ef</sup>	94.50 <sup>ab</sup>
9	22.41 <sup>cdef</sup>	26.67 <sup>de</sup>	93.66 <sup>ab</sup>
10	30.63 <sup>a</sup>	30.63 <sup>a</sup>	96.20 <sup>ab</sup>
11	23.68 <sup>bcd</sup>	27.94 <sup>bc</sup>	96.53 <sup>a</sup>
12	24.41 <sup>bcd</sup>	28.21 <sup>bc</sup>	96.53 <sup>a</sup>
13	11.62 <sup>h</sup>	24.13 <sup>h</sup>	96.53 <sup>a</sup>
14	18.09 <sup>fg</sup>	25.67 <sup>ef</sup>	95.20 <sup>ab</sup>
15	25.15 <sup>bcd</sup>	28.39 <sup>bc</sup>	96.07 <sup>ab</sup>
PROC GLM statistical coefficients			
RMSE*	1.766062	0.377929	1.7196780
R <sup>2</sup>	0.916744	0.970383	0.553208
P values			
DM	< 0.0001	0.0895	< 0.0001
pH	0.0002	< 0.0001	0.1779
S	< 0.0001	< 0.0001	0.9825
DM×pH	< 0.0001	< 0.0001	0.8377
DM×S	0.0109	< 0.0001	0.1449
pH×S	0.0416	0.0065	0.9604
Intercept	0.1001	0.0005	0.2504

<sup>abcde</sup> Indicate significant differences between samples ( $p \leq 0.05$ ). \*RMSE—Root Mean Squared Error.

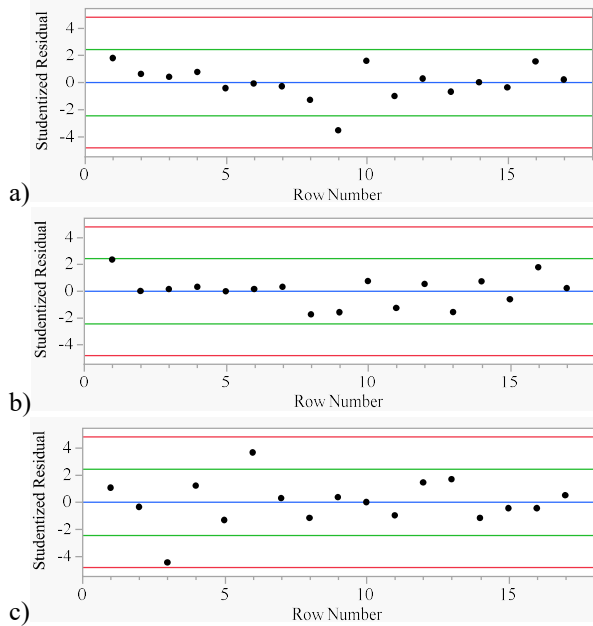
### 3.2 Modelling and optimization

The purpose of drying the coagulant is to reduce the moisture content in order to produce a stable product. Reducing the drying cost and the adverse effects of high temperatures on the product composition can be achieved by reducing the initial moisture content before the drying process. This can be achieved by pressing the coagulant and separating the serum from it, which is mainly water. After the pressing process, the dry matter content (DMC) in the coagulant increases and the moisture content decreases accordingly.

The pressing efficiency (PE), dry matter content (DMC), and yield were significantly ( $p \leq 0.05$ ) influenced by the processing conditions across all 15 design points (DPs) (Table 2). PE exhibited the greatest variability, ranging from 11.62% (DP13) to 30.63% (DP10). It had multiple distinct statistical groupings, indicating strong sensitivity to the experimental factors and their interactions. All three parameters and their paired interaction influenced the PE ( $p \leq 0.05$ ). The highest PE was evaluated in DP10, followed by DP5 and DP7, showing the significant ( $p \leq 0.05$ ) influence of pH (Fig.5). According to Geng et al. (2019) pH value has a great effect on the interaction between PEG and protein in aqueous solutions. Generally, the net surface charge of proteins positively correlates with their solubility, and the proteins are more likely to be precipitated nearing isoelectric point of the solution.

The Bonferroni limits for PE (Fig. 3 a) showed that only one of the studied design points is characterized by a value over minus 2, meaning a strong rejection of null hypothesis. In the same time, the response surface methodology (RSM) (Figure 4) with high lack of fit ( $\geq 0.4286$ ) showed a great model fit (Table 3).

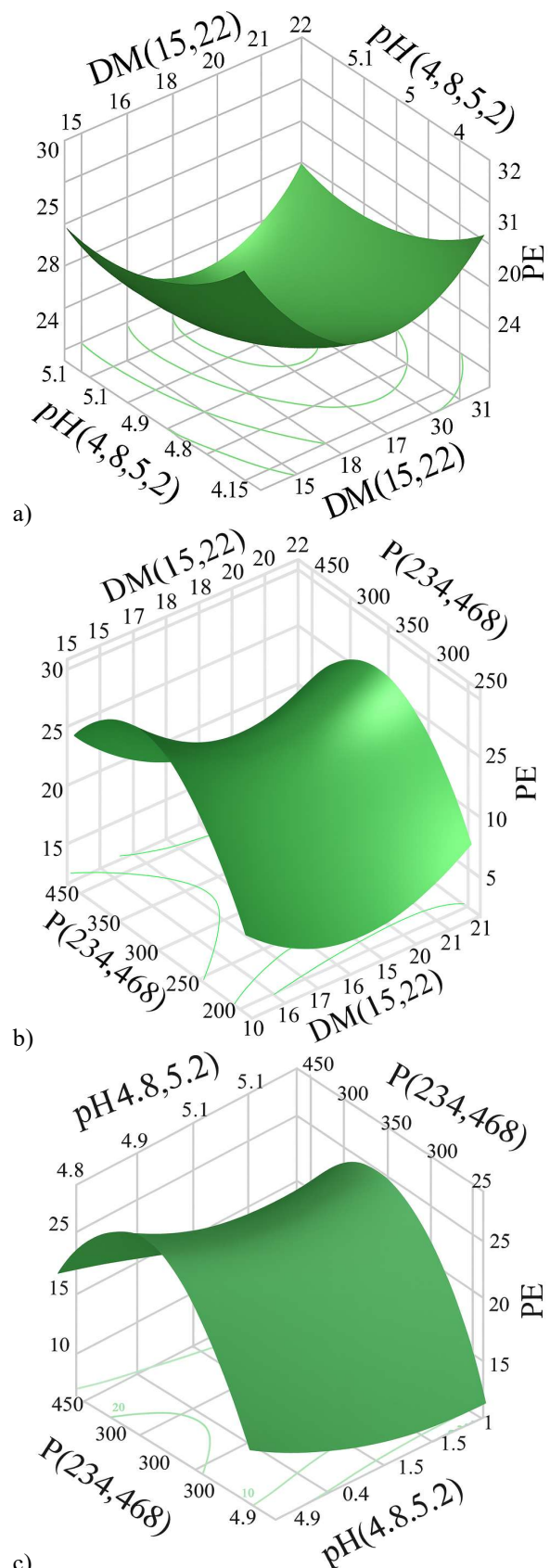
The highest influence on PE was exerted by the pressing force of 351 g/cm<sup>2</sup> (Fig 4 b) and c). With increasing pressing force, PE increases until reaching 300-350 kg and then decreases at a value of 400 kg (Fig 5 b) and c). At this pressure, the PE value is not statistically distinguishable in the pH range 4.8-5.2 (Table 2).



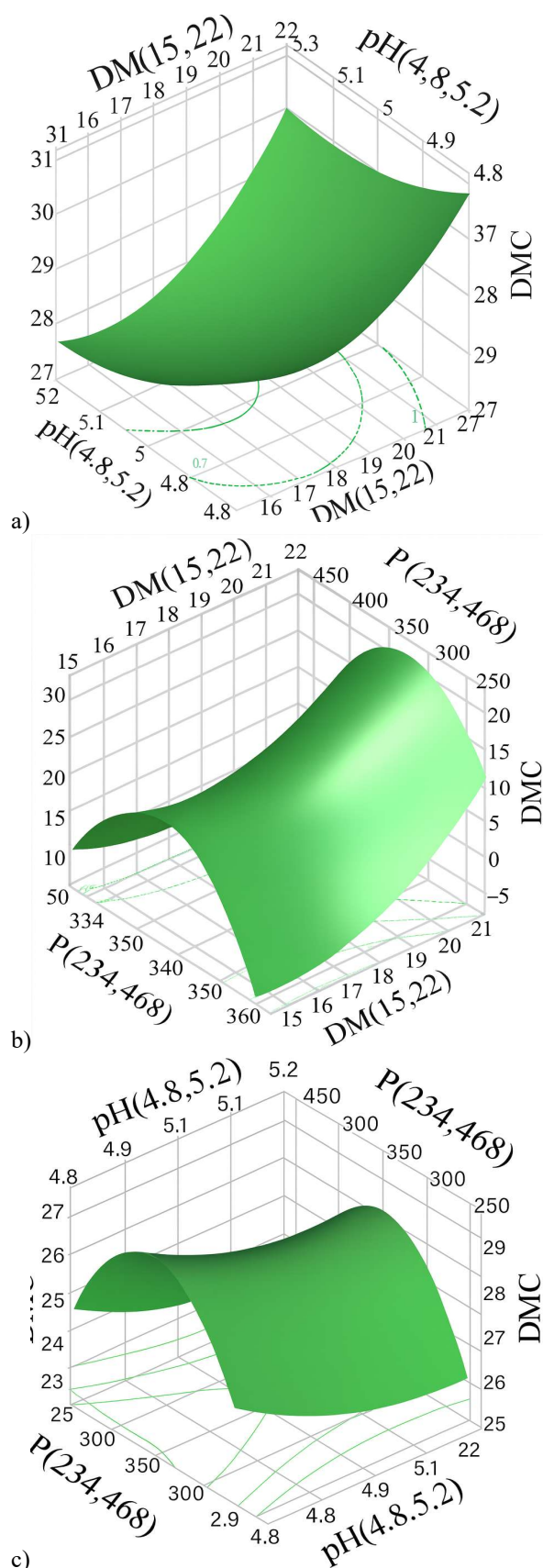
**Fig 3.** Externally studentized residuals with 95% simultaneous limits (Bonferroni) in red, individual limits in green (minus 2)

The other factor studied is the dry matter content of the coagulant. Similarly, to PE, DMC varied from 24.13 to 30.63%, with DP10 and other high-performing treatments clustering in the upper statistical groups (Table 2). This factor is directly related to the final drying process, because the higher the DMC, the more efficient the drying will be. Response surfaces Fig 5 a) shows that with increasing DM and decreasing pH, DMC increases, reaching a maximum value at pH 4.8 and DM 22%.

The Bonferroni limits for DMC (Fig. 3 b) showed that all of the studied design points are fit within the minus 2 limit, rejecting the null hypothesis. In the same time, the RSM (Figure 5) with high lack of fit ( $\geq 0.5725$ ) and  $R^2$  75.64% for DMC showed a great model fit (Table 3). The highest values of DMC are at the medium pressure value 351 g/cm<sup>2</sup>, with the maximum measured value of 30.63% being recorded at DM 22% (Fig. 5 b). It can be seen that the results for DMC are statistically distinguishable and with increasing TA concentration, the dry matter in the coagulant after pressing also increases.



**Fig 4.** Response surfaces of the functions: a) PE, % value =  $f(x_1, x_2)$ , P= 351 g/cm<sup>2</sup>; b) PE, % =  $f(x_1, x_3)$ , DM=18.5% c) PE, % =  $f(x_2, x_3)$ , pH=5.0



**Fig. 5** Response surfaces of the functions: a) DMC, % value =  $f(x_1, x_2)$ ,  $P = 351 \text{ g/cm}^2$ ; b) DMC, % =  $f(x_1, x_3)$ ,  $\text{pH} = 5.0$ ; c) DMC, % =  $f(x_2, x_3)$ ,  $\text{DM} = 18.5\%$

The highest DMC values are at  $351 \text{ g/cm}^2$  P, with no significant influence of pH (DPs 11,12 and 15) (Table 2, Figure 5 c).

PE and DMC are similar indicators, with the first showing the percentage of serum extracted during pressing, and the second the dry matter in the coagulant after pressing. It can be concluded that with increasing DM and decreasing pH to 4.8, DMC also increases, with DM being a more significant ( $p \leq 0.05$ ) indicator. Also, average pressure values of  $351 \text{ g/cm}^2$  are most favourable for the pressing process, due to clogging of the holes at higher pressures and inability for serum to pass through.

The GLM analysis confirmed these observations: DMC were strongly affected by DM, pH, P, and their two-way interactions ( $p \leq 0.05$ ), while only DM significantly influenced yield. The high coefficients of determination for PE ( $R^2 = 0.92$ ) and DMC ( $R^2 = 0.97$ ) indicate excellent model fit, whereas yield exhibited lower explanatory power ( $R^2 = 0.55$ ). Overall, the results show that PE and DMC are highly responsive to processing parameters, particularly under the conditions represented by DP10, whereas yield remains relatively stable across treatments.

In contrast, yield showed a narrower range (91.32–96.76%) and fewer significant ( $p \leq 0.05$ ) differences among DPs (Table 2). The GLM combined with the RSM plots showed that only DM of the raw material had an influence ( $p \leq 0.05$ ) on the yield (Fig. 6).

The Bonferroni limits for Yield (Fig. 3 c) showed that two of the studied design points are characterized by a value over plus or minus 2, meaning a strong rejection of null hypothesis. This combined with the response surface methodology (RSM) (Figure 6) with high lack of fit ( $\geq 0.1876$ ) showed a great model fit (Table 3). It can be seen from Figure 6 a) at  $P = 351 \text{ g/cm}^2$  the yield increases constantly with increasing DM. Figure 6 b) shows the interaction of pH and P at DM - constant (18.5%). The maximum yield values are at pH 4.8 and P  $351 \text{ g/cm}^2$ . Three-dimensional response surface chart at pH 5.0 (Figure 6 c) showed a significant ( $p \leq 0.05$ ) influence of the DM factor as the yield continuously increased until reaching a maximum value at 22%. Therefore, the optimum conditions for yield were identified for obtaining a dry product from TA.

Pressing is a widely used method in food processing, in which, as a result of applied force, a liquid fraction is formed, which passes through the holes of the sieve, and a solid fraction, which remains in the basket. The efficiency of pressing depends on the raw material (liquid content and particle size distribution), as well as on the conditions of pressure, time, etc. [28].

A potential clogging of the sieve holes or compaction of protein aggregates at high pressure and the inability of the liquid to pass can be explanation of the evaluated phenomenon.

We can summarize that with increasing DM, the protein content increases, and therefore its protein-protein interactions. This causes the formation of more aggregates than at low DM values, accompanied by the separation of their hydration shell, which increases the liquid fraction.

The second most important factor is the concentration of dry matter in the raw material (Fig 4 a), b) with proteins accounting for the largest share.

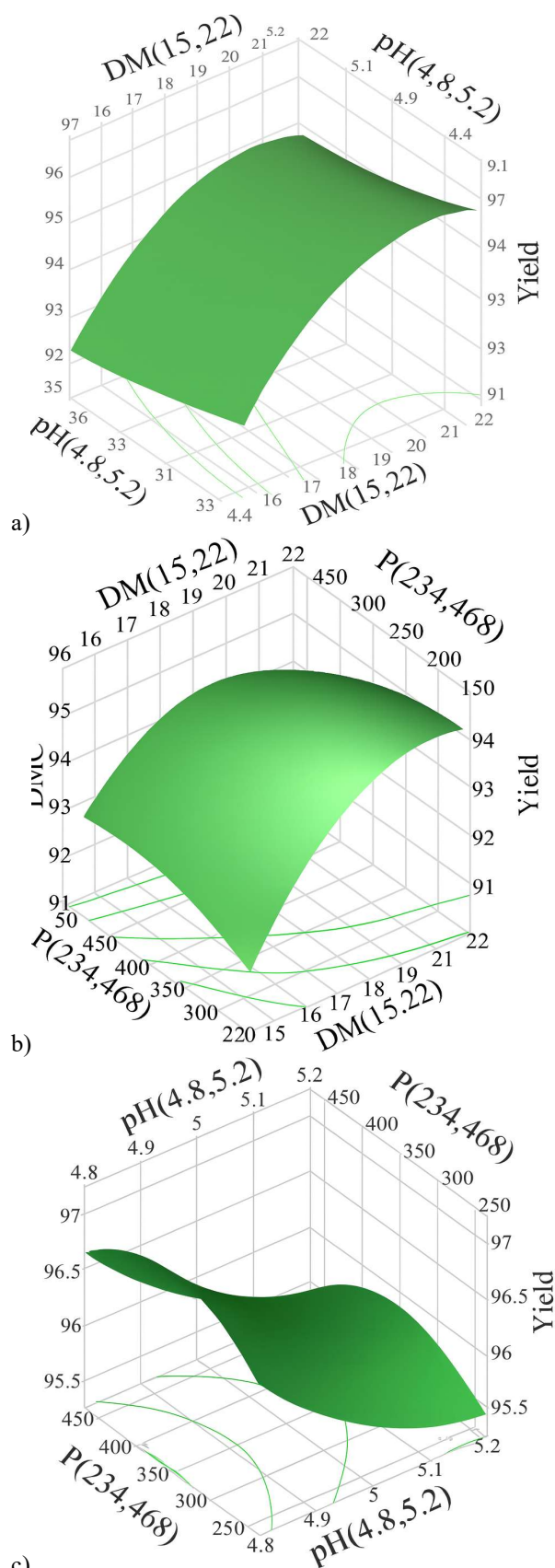


Fig 6. Response surfaces of the functions: a)  $Y, \% = f(x_1, x_2)$ ,  $P = 351 \text{ g/cm}^2$ ; b)  $Y, \% = f(x_1, x_3)$ ,  $S = 18.5\%$ ; c)  $Y, \% = f(x_2, x_3)$ ,  $pH = 5.0$

Proteins are polymers of hundreds of amino acids linked together by peptide bonds to form peptide chains. The side chains and functional groups interact with each other through very weak interactions involving Van der

Vaal's forces, hydrogen bonds, electrostatic and hydrophobic interactions, and the final effect is the native structure of the protein [29]. In an aqueous solution such as technical protein, the dipoles of water are located mostly around the hydrophilic groups on the surface of the protein, forming a hydration shell through hydrogen bonds with the amino acid residues and between the water molecules themselves in the layer.

Thermal denaturation leads to unfolding of the protein chain, exposing the hydrophobic centres to the surface, causing destabilization of the hydration shell [30]. At pH values close to the isoelectric point and at high ionic strength, the charge strength between protein molecules is quite low, leading to the formation of clusters of aggregates [31]. Along with aggregation of protein molecules, the low charge near the *pI* leads to low hydrophilicity causing syneresis [32]. Syneresis is the term used to describe the loss of liquid from food systems due to changes in the physicochemical conditions of the system itself. The volume or weight of liquid loss through syneresis is measured directly by its volume or by changes in the weight of the remaining solids. Syneresis leads to a concentration of the solid fraction because the liquid is mostly water [33]. According to Yu et al. [22] the pH-shifting can lead to molecular structural modification of egg white protein as well as improving emulsifying capacity and stability. In the same time, Khemakhem et al. [34] used citric acid to correct the pH egg white before thermal treatment leading to significant decrease of water holding capacity of obtained gels. They hypothesised that due to the fact ovalbumin is the most abundant protein in egg albumen with *pI* 4.5, where the electrostatic repulsions are reduced, and the net charge approaches zero as well as electrostatic interactions between protein and water were reduced. The same authors also report a significant influence of pH on the texture profile of the egg white gels. The sample with pH close to the isoelectric point were cauterized by decreased hardness and elasticity, compared to the control.

On the other hand, the presence of egg yolk proteins in TA such as  $\gamma$ -Livetin,  $\alpha$ -Livetin (LDL), Apovitellin 3+4 and Apovitellin 8 (HDL) (Figure 2), the first ones known for their excellent emulsifying properties can possibly explain our findings. For example, in DPs 1, 5 and 7 the DM of raw material was diluted with water to 15%. Those DPs had a significantly better PE ( $p \leq 0.05$ ), compared to DPs 2, 6 and 8, respectively with 22% DM. The higher total concentration of egg yolk proteins potentially forms a much more stable emulsion leading to more strong water binding and decreased water separation during pressing.

The loss of yield in this process is due to the presence of soluble substances in the liquid fraction separated during pressing. A high dry matter content implies a lower water content, respectively a lower possibility of extract passing into the serum.

Despite the fact that all three studied factors (DM, pH and P) influenced PE ( $p \leq 0.05$ ), as well as The Summarized effect of RSM for the three evaluated parameters, showed that DM ( $p \leq 0.00059$ ) and  $P^2$  ( $p \leq 0.01177$ ) had a significant influence (Fig 7.)

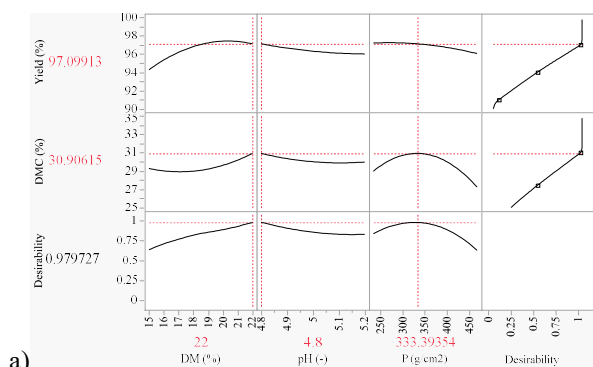
**Table 3.** Summary of Fit of RSM

	PE, %	DMC, %	Yield, %
R <sup>2</sup>	0.681227	0.756375	0.900882
RMSE	4.193063	1.324243	0.73218
Mean of Response	22.33647	27.21	95.30588
Lack of Fit	0.4286	0.5725	0.1876

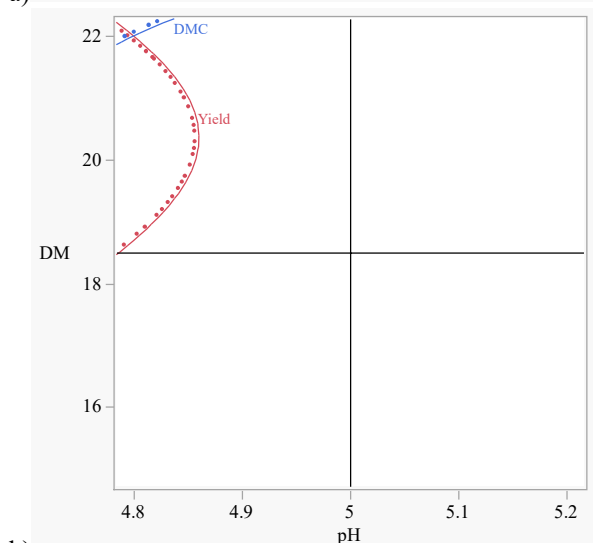
Source	Logworth		PValue
DM(15,22)	3.226		0.00059
P*P	1.929		0.01177
DM*DM	1.710		0.01951
pH(4.8,5.2)	1.429		0.03725
DM*P	1.277		0.05281
P(234,468)	0.842		0.14400
pH*pH	0.236		0.58085
pH*P	0.207		0.62061
DM*pH	0.191		0.64367

**Fig. 7.** Summarized effect of RSM

The mathematically defined combined optimum (Fig. 8) according to the three target functions (PE, DMC and Yield) was determined at: 22% DM ( $x_1$ ), 4.80 pH ( $x_2$ ) and 333.3954 g/cm<sup>2</sup> P ( $x_3$ ).



a)



b)

**Fig 8.** Optimization of the processing parameters: a) Desirability of the model; b) Contour profile of the optimal response

This research was funded by the Bulgarian National Science Fund, grant number KP-06-M76/2, from 5 December 2023, entitled: Opportunities for utilizing waste from egg processing.

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