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Evaluation of the rheological characteristics of processed cheese produced from sodium caseinate, pea protein and corn oil

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ABSTRACT

In this research, the effects of sodium caseinate, pea protein, and corn oil on oiling off, stickiness, Work of adhesion, and Work of shear process cheese were investigated. The results were modeled and analyzed using the response surface method in the form of central composite design. Optimization of processed cheese formulation using central compound design for three independent variables including pea protein powder (0, 7.5, 15%W/W), corn oil (0, 8, 16%W/V) and sodium caseinate (0, 2.5, 5%W/W) was done. In general, the effect of all variables on oil return characteristics, stickiness, Work of adhesion and Work of shear were significant. Based on the obtained results, the oiling off cheese samples increased with the increase of corn oil levels. In the corn oil/sodium caseinate interaction, with the increase in the percentage of corn oil and sodium caseinate, the Work of adhesion of the cheese samples decreased significantly. In the interaction of pea protein/sodium caseinate, with the increase in the percentage of pea protein, the work required to cut the cheese samples increased significantly. In the interaction of pea protein/sodium caseinate, with increasing the percentage of pea protein, the work required to cut the cheese samples increased significantly; while the addition of sodium caseinate led to a decrease in the studied parameter. Therefore, sodium caseinate, pea protein, and corn oil are good supplements to reduce each other's negative effects on the properties of oil return, expandability, and the work required to cut enriched processed cheese, and the simultaneous use of these three in processed cheese ultimately improves the properties of processed cheese. It was mentioned in comparison with the witness sample. The results of this study are favorable for the development of cheese and ultra-profitable dairy products containing sodium caseinate, pea protein and corn oil.

1.Introduction

Cheese, as a valuable food source, is considered one of the oldest and most important dairy products. Due to its ease of consumption, either alone or in combination with other foods, it has gained great popularity and high demand [1]. However, the cheese-making process is time-consuming, requires a considerable amount of energy, and, along with the rising cost of milk, has led to cheese becoming an expensive product. Moreover, cheese contains high levels of cholesterol and saturated fat, which are associated with an increased risk of cardiovascular disease. Therefore, the development of a plant-based cheese product that can mimic the unique characteristics of natural cheese, such as stretchability and stringiness upon melting, has attracted the attention of researchers [2]. Processed cheese is produced by blending natural cheese with emulsifying salts (such as sodium phosphates and citrates), along with other dairy and non-dairy ingredients, followed by heating and continuous mixing to form a homogeneous product with extended shelf life [3]. The emulsifying salts, when combined with heating and shearing, break the calcium phosphate bridges, enabling the para-casein molecules to associate with one another. As a result, soluble casein forms a network structure responsible for the viscosity of processed cheese. During heating, para-caseinate emulsifies fat, producing a stable oil-in-water emulsion—a process referred to as creaming. This process yields a homogeneous product with prolonged shelf stability; however, due to the addition of emulsifying salts (sodium-based), processed cheese typically contains relatively higher sodium levels compared to hard cheeses [4]. Sodium caseinate is produced by adding sodium hydroxide to acid casein and is essentially a mixture of casein monomers and small particles formed after the removal of colloidal calcium phosphate from casein micelles [5]. In processed food products, sodium caseinate is widely used as a protein source due to its physicochemical, nutritional, and functional properties. Nowadays, because of its ability to form protein–ligand complexes and casein micelles, its applications have expanded to include stabilizers, encapsulation, emulsification, edible films and coatings, and other food-related uses [6]. Sodium caseinate is commonly employed in baked goods, breakfast cereals, meat products, desserts, puffed snacks, and processed cheese [7]. Due to its amphiphilic nature and disordered structure, caseins can rapidly adsorb onto the surface of oil droplets, forming a dense, entangled layer that protects newly formed droplets from flocculation. Compared to proteins such as soy protein and whey protein, corn oil emulsions stabilized with sodium caseinate exhibit greater oxidative stability at pH 3. The incorporation of sodium caseinate in processed cheese production results in higher pH, lower firmness, higher degree of fat emulsification, and greater casein dissociation. Sodium caseinate is also used as a partial protein substitute in cheese, contributing similarly to the formation and stabilization of processed cheeses [8]. Pea protein consists of four main fractions (globulins, albumins, prolamins, and glutelins), among which globulins and albumins are the primary storage proteins in pea seeds. Pea protein possesses a balanced amino acid profile with a high lysine content. Its composition and structure, as well as processing conditions, significantly influence its physicochemical properties such as hydration, rheological behavior, and surface characteristics. Due to its availability, low cost, nutritional value, and health benefits, pea protein can serve as a novel and effective substitute for animal proteins in various food applications [9]. Moreover, the use of pea protein in food systems has been investigated for applications such as encapsulation of bioactive compounds, edible films, extruded products, and as a replacement for cereal flours, fats, and animal proteins [10]. Maize (*Zea mays* L.) is a herbaceous plant and one of the most important cereal crops, ranking third globally in terms of cultivated area. Among vegetable oils, corn oil ranks tenth in annual production, accounting for about 2% of total global oil output [11]. Corn kernel oil is slightly yellow in color with a mild and characteristic flavor. It is a valuable source of bioactive lipids such as phytosterols, tocopherols, tocotrienols, and carotenoids. Compared with other vegetable oils, corn oil exhibits relatively higher nutritional quality and oxidative stability [12]. Corn oil is also categorized among vegetable oils rich in linoleic and oleic acids. Approximately 60%, 25%, and 15% of its fatty acids are polyunsaturated, monounsaturated, and saturated, respectively. Among unsaturated fatty acids, corn oil contains on average 30.5% oleic acid (C18:1), 52% linoleic acid (C18:2), and only 1% linolenic acid (C18:3). Similar to other vegetable oils, the fatty acid composition of corn oil depends on factors such as genotype, climatic conditions, and growing season [13]. Several related studies have been conducted in the field relevant to the present research. For instance, Yang et al. (2021) [14] compared the physicochemical properties and flavor compounds of yogurts based on pea protein and mung bean protein. Their results indicated that mung bean protein could be utilized to produce plant-based yogurt alternatives with better quality than those prepared with pea protein. Similarly, Narala et al. (2022) [15] investigated the use of inulin as a fat replacer in pea protein-based plant ice cream and its effects on textural and sensory attributes. Their findings showed that increasing inulin content in the formulation prolonged the first dripping time of ice cream. Significant differences were also observed in hardness and adhesiveness of pea protein ice cream depending on the inulin content in the formulation. The aim of the present study is to evaluate the rheological properties of processed cheese produced using sodium caseinate, pea protein, and corn oil, in order to achieve an optimized formulation for developing a health-promoting product with high quality and extended shelf life.

2. Materials and Methods

2.1.Materials

For the experiments, Feta cheese with the following specifications was used: pH 4.99, acidity 1.3%, moisture 62.2%, fat 15%, protein 13.18%, and salt 2.85%. Pea protein was obtained from the OEM/ODM Company (China) with the following characteristics: moisture < 8%, total bacterial count < 10 Wcfu/g, ash 5.6%, protein > 80%, and solubility: good. Refined corn oil was purchased from Ladan Company (Iran). In addition, SS90 emulsifying salt (sodium polyphosphate) was obtained from CORINO Company (Thailand).

2.2. Preparation of Processed Cheese

To prepare processed cheese, the dry matter and fat content of the base cheese (Feta cheese) were first determined. A specific amount of base cheese was then weighed, and according to its fat-free dry matter content, predetermined percentages of pea protein powder (0, 7.5, and 15%) and corn oil (0, 8, and 16%) were added. The mixture was blended at near ambient temperature for 1–2 hours until a completely soft and homogeneous paste was obtained. Subsequently, sodium caseinate emulsifying salt (0, 2.5, and 5%) was thoroughly mixed with the other ingredients and transferred to the cooking apparatus. The mixture was stirred without applying shear stress until the temperature reached 70–75 °C. Thereafter, mixing under shear stress (at 3000 rpm) was performed at 85 °C for 4–5 minutes. After the specified time, the product was discharged and hot-filled into 150 g containers, followed by immediate cooling in a cold room for 3–4 hours until the temperature reached 5–10 °C [16].

3.2. Oil Leakage Measurement

To determine meltability and oil leakage, an image processing method was employed. Cheese samples with a thickness of 5 mm and a diameter of 22 mm were placed on plates containing filter paper and photographed using a digital camera (Canon PowerShot A550) at 2× magnification inside a chamber equipped with a 30 W fluorescent lamp, positioned 20 cm above the samples. After the initial photography, the samples were placed in an oven at 95 °C for 20 minutes. Following heating, the samples were removed and photographed again. The captured images were processed using *ImageJ* software to calculate the surface area of the cheese samples before and after oven treatment, as well as the area of the oil halo diffused into the filter paper. Finally, oil leakage of the cheese samples was calculated according to Equation (2-1).

$$POA = (A_f/A_0) \times 100 \qquad \text{Eq.1}$$

A_f is the oiled surface area of the filter paper, A_0 is the initial surface area of the sample

4.2. Adhesion, Spreadability, and Cutting Test

To evaluate the spreadability of cheese samples, a TA.XT Plus texture analyzer (Stable Micro Systems, UK) was used. The processed cheese was placed in the device's sample holder, and a conical probe was driven into the sample at a speed of 3 mm/s. After traveling a distance of 23 mm, the force–time curve was recorded using the instrument software. The analyzed parameters included adhesion, the work required to overcome adhesion (spreadability), and the work required for cutting [17].

3. Statistical Analysis

The optimization of the processed cheese formulation was performed using a central composite design (Table 1) at three levels for three independent variables: pea protein powder (0, 7.5, 15%), corn oil (0, 8, 16%), and sodium caseinate (0, 2.5, 5%). The characteristics of oil leakage, adhesion, spreadability, and cutting of the cheese samples were analyzed using analysis of variance (ANOVA) in Design Expert 11 software. To evaluate the validity of the predicted models, tests such as lack of fit, coefficient of variation (CV%), R^2, and adjusted R^2 (Adj-R^2) were conducted. The significance level of the factors was considered at 95%.

Table 1: Experimental design of response surface in the form of central composite design

Run	%Corn Oil	%Pea Protein Powder	%Sodium Caseinate
1	8.00	15.00	**2.50**
2	8.00	7.50	**2.50**
3	8.00	7.50	**2.50**
4	8.00	7.50	**2.50**
5	8.00	7.50	**2.50**
6	8.00	7.50	**2.50**
7	8.00	7.50	**5.00**
8	0.00	15.00	**0.00**
9	16.00	0.00	**0.00**
10	0.00	0.00	**5.00**
11	0.00	0.00	**0.00**
12	8.00	7.50	**0.00**
13	16.00	15.00	**0.00**

14	0.00	7.50	**2.50**
15	8.00	0.00	**2.50**
16	0.00	15.00	**5.00**
17	16.00	15.00	**5.00**
18	16.00	7.50	**2.50**
19	16.00	0.00	**5.00**

4. Results and Discussion

4.1. Oil Leakage of Cheese

The oil leakage of processed cheese samples containing different levels of corn oil, pea protein, and sodium caseinate is presented in Figure 1. According to Figure 1 and the results of the analysis of variance (Table 2), the individual effects of corn oil, pea protein, sodium caseinate, and the interaction of corn oil/sodium caseinate on oil leakage of processed cheese were significant ($P \leq 0.05$). The results indicated that increasing the levels of corn oil led to higher oil leakage, whereas the addition of sodium caseinate at different levels resulted in a decrease in oil leakage.

As reported in previous studies, vegetable oils dispersed within the cheese matrix can disrupt the protein network, leading to the formation of a weak emulsion and the release of oil from the protein structure, thereby increasing oil leakage [18]. Furthermore, increasing the levels of sodium caseinate in the processed cheese formulation protects the emulsion due to its surface-active properties. During denaturation, the formation of nonpolar groups creates a protective layer that prevents emulsion breakdown and inhibits the aggregation and release of fat particles from the final product [16].

Ahmed et al. (2019) [19] reported that high levels of flaxseed oil, used as a butter substitute in processed cheese, resulted in increased oil leakage. Teng et al. (2019) [20] also demonstrated that the addition of various vegetable oils (corn, sunflower, rice bran, and coconut) in processed cream cheese led to increased oil leakage, particularly in samples containing sunflower oil.

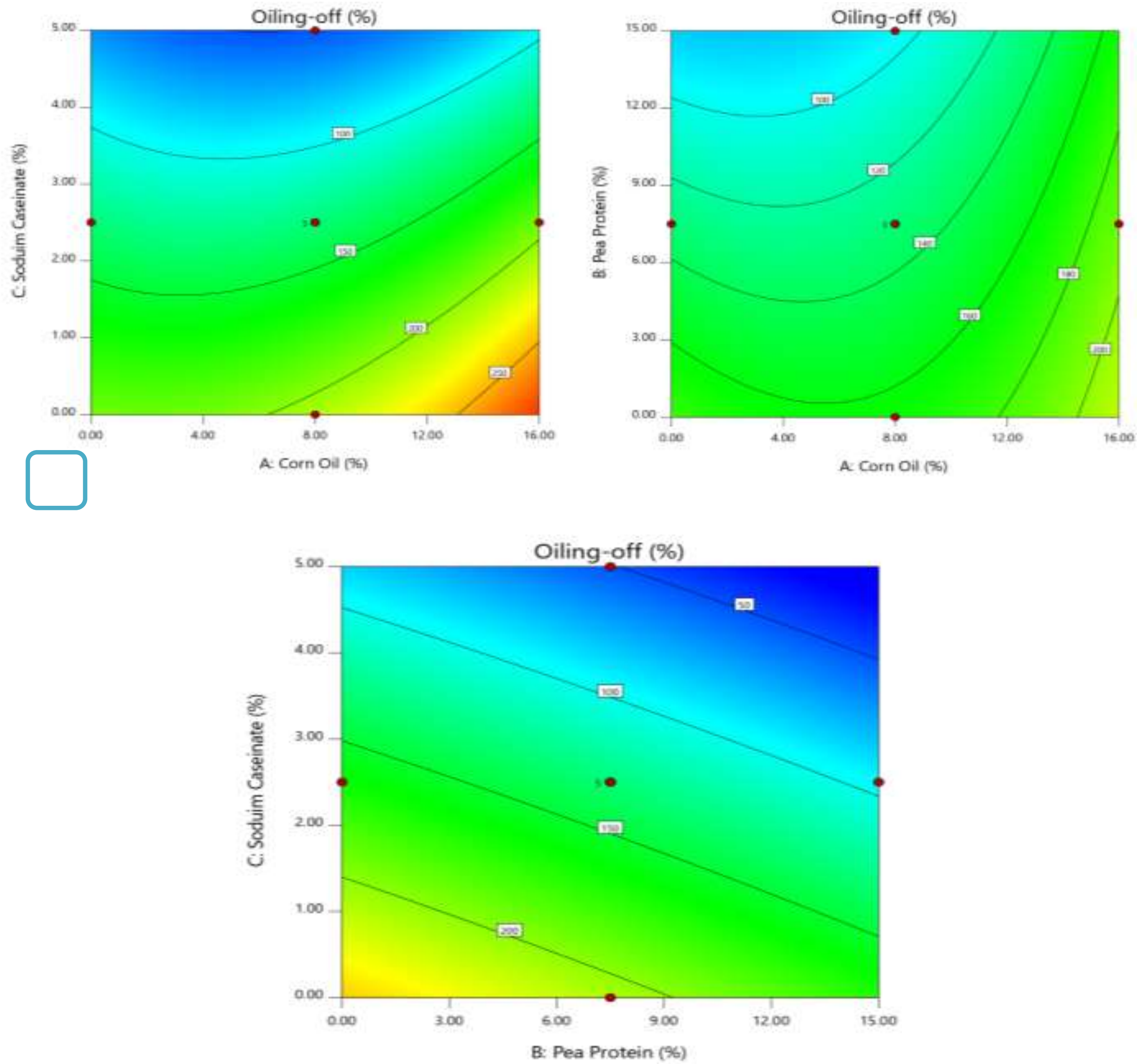

Fig 1: (a, b, c) Counterplot effect of pea protein, corn oil and sodium caseinate on the oiling - off processed cheese

Table 2: Regression coefficient of Oiling - off in processed cheese

Factor	Oiling – off (%)
Model	**10061.08****
Intercept	**239.36**
A- corn oil	**-3.28****
B- Pea protein	**-6.08****
C- Sodium caseinate	**-24.14****
AB	**0.2**
AC	**-0.8***
BC	**0.03**
A^2	**0.47***

B^2	**-0.02**
C^2	**-0.23**
R^2	**0.97**
Adj-R^2	**0.94**
Lack of Fit	**2.87^{ns}**
%C.V	**0.76**

p≥0.05 :ns(non-significant) ،p<0.01:** ،p≤0.05:*

4.2.Work Required to Overcome Adhesion

According to the analysis of variance results (Table 3), the individual effects of corn oil, pea protein, and sodium caseinate, as well as the interaction between corn oil and sodium caseinate, on the work required to overcome adhesion in processed cheese were significant (P ≤ 0.05). As shown in Figure 2, in the interaction of corn oil and sodium caseinate, increasing the percentages of both corn oil and sodium caseinate significantly decreased the work required to overcome adhesion in cheese samples. Conversely, the interactions of corn oil/pea protein and pea protein/sodium caseinate did not show statistically significant effects on this parameter (P > 0.05). The reduction in the work required to overcome adhesion can be attributed to a decrease in the force needed to overcome the attraction between the product surface and the probe, due to reduced product stickiness. Additionally, it may be related to the formation of a weaker gel by the components used in the cheese formulation [21]. Similar results regarding the reduction of adhesion and work required to overcome adhesion were reported by Giri et al. (2014b) [22], who observed that the addition of inulin as a fat replacer and a high-nutritional-value dietary fiber in processed spreadable cheese decreased adhesion. The predictive model used was significant, with a non-significant lack of fit, R^2 and adjusted R^2 (Adj-R^2) of 0.99 and 0.98, respectively, and a low coefficient of variation (CV), indicating the efficiency and reliability of the model.

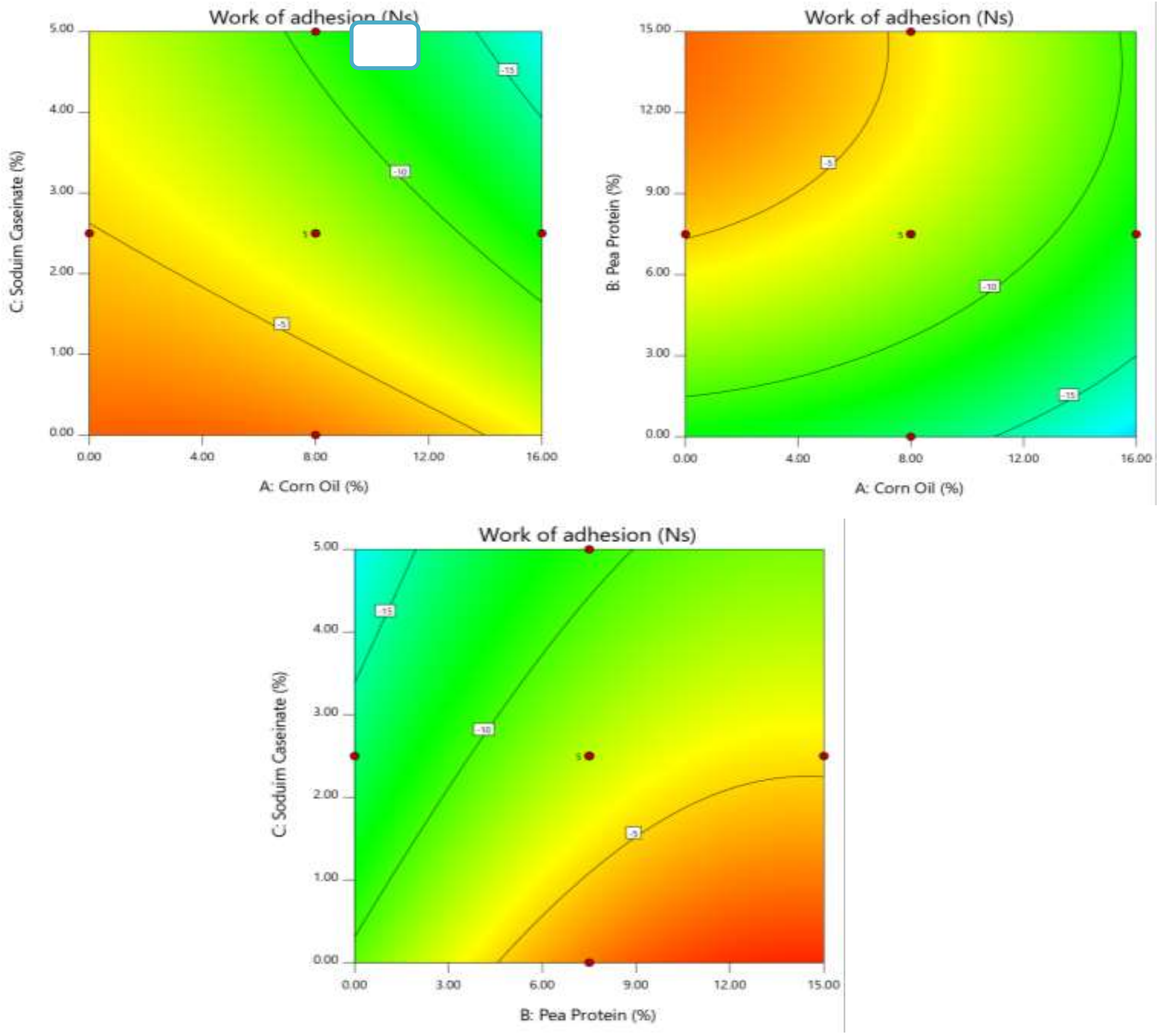

Fig 2: (a, b, c) Counterplot effect of pea protein, corn oil and sodium caseinate on the Work of adheion processed cheese

4.3. Work Required for Cutting

According to the analysis of variance results (Table 3), the individual effects of corn oil, pea protein, and sodium caseinate, as well as the interaction between pea protein and sodium caseinate, on the work required for cutting processed cheese were significant ($P \leq 0.05$). As shown in Figure 3, in the interaction of pea protein and sodium caseinate, increasing the percentage of pea protein significantly increased the work required for cutting the cheese samples, whereas the addition of sodium caseinate led to a reduction in this parameter.

This phenomenon can be explained as follows: in addition to the added pea protein, casein protein is already present in the processed cheese structure. The protein–protein interactions result in a compact and dense structure, thereby increasing the work required for cutting. However, excessive protein content can weaken the protein network; thus, the addition of sodium caseinate reduces protein–protein interactions within the texture, leading to a decrease in cutting work (Figure 3) [23]. Conversely, the interactions of corn oil/pea protein and corn oil/sodium caseinate did not have statistically significant effects on the cutting work ($P > 0.05$). The predictive model used was significant, with a non-significant lack of fit, R^2 and adjusted R^2 (Adj-R^2) values of 0.97 and 0.95, respectively, and a low coefficient of variation (CV), indicating the reliability and efficiency of the model.

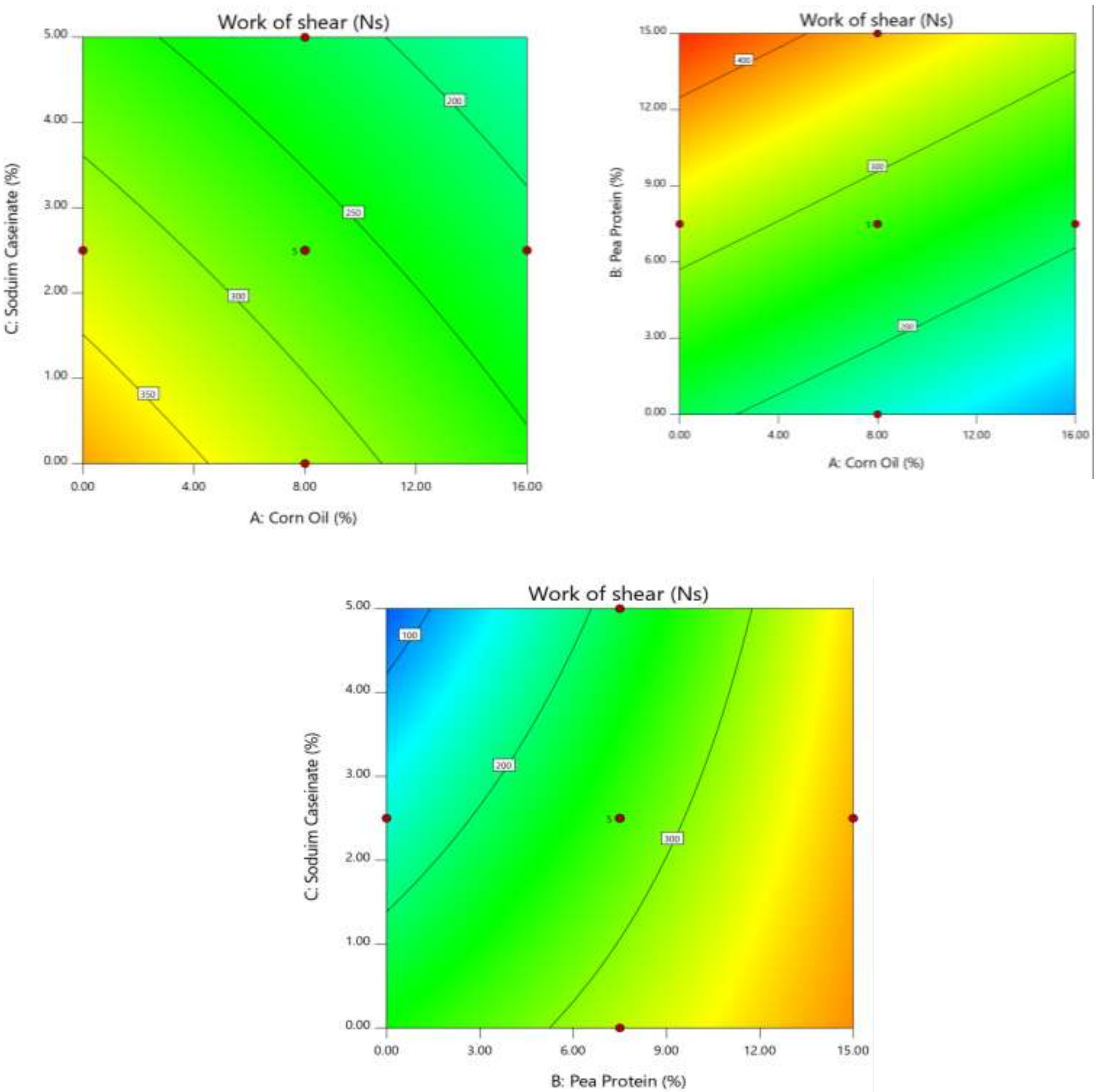

Fig 3: (a, b, c) Counterplot effect of pea protein, corn oil and sodium caseinate on the Work of shear processed cheese

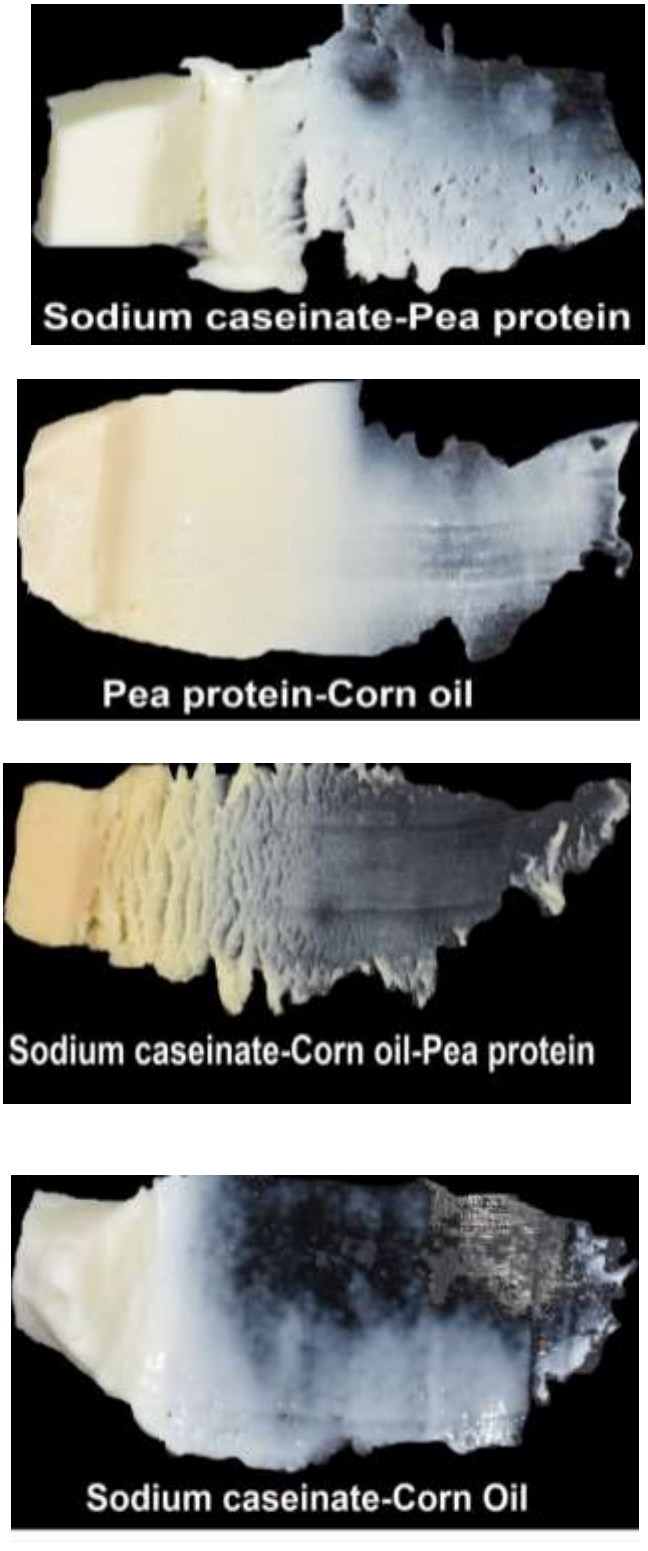

Fig 4: A view of the shear stress of processed cheese tissue containing the interactions of corn oil, pea protein and sodium caseinate.

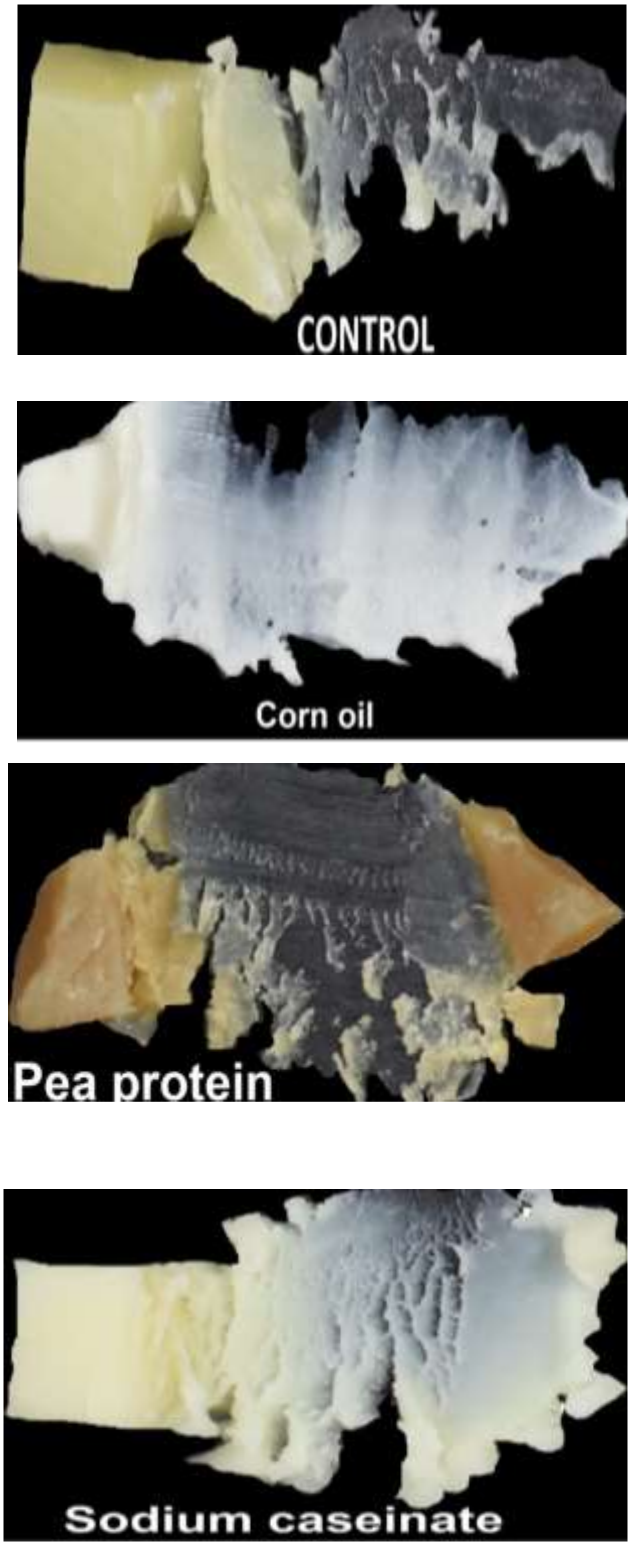

Fig 5: A view of the shear stress of the cheese texture of the control process, with corn oil, pea protein and sodium caseinate

4.4.Adhesion

According to Table 3 and the results of the analysis of variance, the individual effects of corn oil, pea protein, sodium caseinate, and the interaction between corn oil and sodium caseinate on the adhesion of processed cheese were significant ($P \leq 0.05$). As shown in Figure 6, in the interaction of corn oil and sodium caseinate, increasing the percentage of added corn oil resulted in a reduction of cheese sample adhesion. The addition of sodium caseinate did not cause a significant change in adhesion. This phenomenon can be attributed to the degree of interaction between the protein matrix and the lipid phase; as the corn oil content increases, the interaction between the two phases decreases, leading to reduced adhesion [24].

Similar results regarding the reduction of adhesion were reported by Szafrańska & Sołowiej (2020) [25], who observed that adding various fibers (bamboo, acacia, potato, or citrus) as fat replacers in acid-casein-based processed cheese sauces reduced adhesion. Conversely, the interactions of pea protein/corn oil and pea protein/sodium caseinate did not have statistically significant effects on adhesion ($P > 0.05$).

The predictive model used was significant, with a non-significant lack of fit, R^2 and adjusted R^2 (Adj-R^2) values of 0.94 and 0.91, respectively, and a low coefficient of variation (CV), indicating the efficiency and suitability of the obtained model.

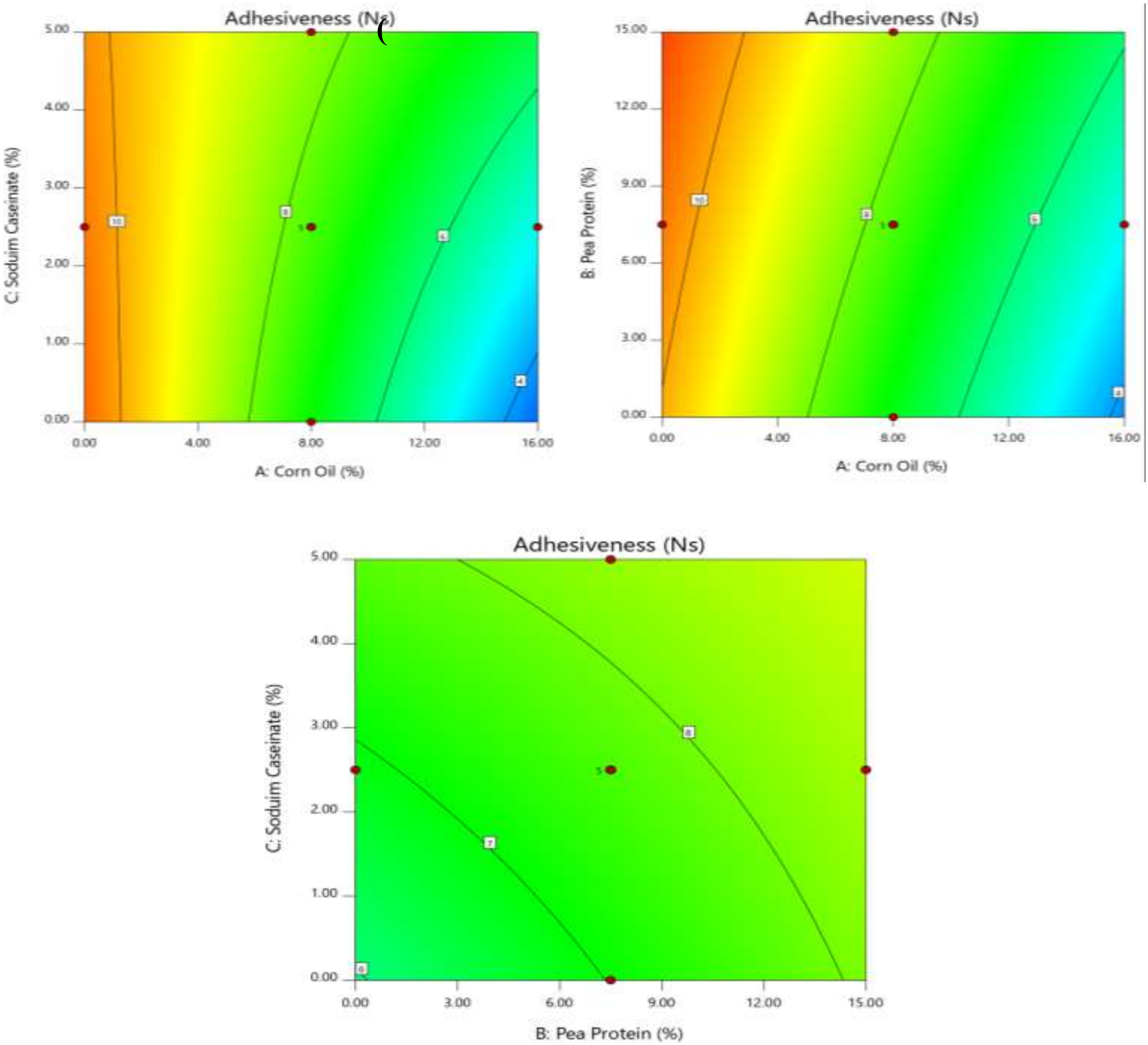

Fig 6: (a, b, c) Counterplot effect of pea protein, corn oil and sodium caseinate on the Adhesiveness processed cheese

Table 3: Regression coefficients of the extensibility properties of processed cheese

Factor	Adhesiveness	Work of adhesion	Work of shear
Model	1109.27**	56.15**	**3155.43****
Intercept	-22.02	-9.16	**311.35**
A- corn oil	-0.15**	0.1**	**-7.82****
B- Pea protein	-2.35**	1.2**	**9.97****
C- Sodium caseinate	-6.45**	-1.18**	**-38.24****
AB	0.09*	$-6.87\times10^{-0.03}$	**-0.02**

AC	0.3^{*}	-0.08^{**}	**0.37**
BC	-0.24	$8\times10^{-0.04}$	**1.91^{**}**
A^2	0	-0.01	**0**
B^2	0	-0.03^{**}	**0**
C^2	0	-0.06	**0**
R^2	0.93	0.99	**0.97**
Adj-R^2	0.89	0.98	**0.95**
Lack of Fit	2.17^{ns}	0.34^{ns}	**4.46^{ns}**
C.V%	0.77	0.94	**0.91**

p≥0.05 :ns(non-significant) ‹p<0.01:** ‹p≤0.05:*

5. Conclusion

In this study, the characteristics of oil leakage, spreadability (adhesion and the work required to overcome adhesion), and cutting work of processed cheese produced from sodium caseinate, pea protein, and corn oil were evaluated. The results indicated that increasing the corn oil content led to higher oil leakage, whereas increasing the sodium caseinate content had the opposite effect. Furthermore, increasing the percentage of corn oil reduced the adhesion of cheese samples, while the addition of sodium caseinate did not result in significant changes in adhesion.
In the interaction between pea protein and sodium caseinate, increasing the pea protein content significantly increased the work required for cutting the cheese samples, whereas adding sodium caseinate reduced this parameter. Moreover, increasing both corn oil and sodium caseinate significantly decreased the work required to overcome adhesion.
Therefore, sodium caseinate, pea protein, and corn oil act as complementary components that mitigate each other's negative effects on oil leakage, spreadability, and cutting work of enriched processed cheese. The simultaneous use of these three ingredients ultimately improved the aforementioned properties compared to the control sample. The findings of this study are valuable for the development of functional dairy products containing sodium caseinate, pea protein, and corn oil.

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مقاله علمی_پژوهشی

ارزیابی ویژگی‌های رئولوژیکی پنیر پروسس تولیدی از کازئینات سدیم، پروتئین نخود و روغن ذرت

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چکیده

در این پژوهش به بررسی اثرات کازئینات سدیم، پروتئین نخود و روغن ذرت بر میزان پس دهی روغن، چسبندگی، کار لازم برای غلبه بر چسبندگی و برش پنیر پروسس تولیدی پرداخته شد. نتایج به روش سطح پاسخ در قالب طرح مرکب مرکزی مدل سازی و تجزیه و تحلیل شدند. بهینه سازی فرمولاسیون پنیر پروسس با استفاده از طرح مرکب مرکزی برای سه متغیر مستقل شامل پودر پروتئین نخود (۰، ۷/۵، ۱۵٪وزنی/ وزنی)، روغن ذرت (۰، ۸، ۱٦٪ وزنی / حجمی) و کازئینات سدیم (۰، ۲/۵، ۵٪ وزنی / وزنی) انجـام شـد. به طور کلی اثر همه متغیرها بر ویژگی های پس دهی روغن، چسبندگی، کار لازم برای غلبه بر چسبندگی و برش پنیر پروسس معنادار بود. با توج به نتایج به دست آمده با افزایش سطوح روغن ذرت میزان پس دهی روغن نمونه‌های پنیر افزایش یافت. در برهمکنش روغن ذرت/ کازئینات سدیم با افزایش درصد روغن ذرت و کازئینات سدیم کار مورد نیاز برای غلبه بر چسبندگی نمونه‌های پنیر به طور معناداری کاهش یافت. در برهکنش پروتئین نخود/ کازئینات سدیم با افزایش درصد پروتئین نخود کار لازم برای برش نمونه‌های پنیر به طورمعناداری افزایش یافت.؛ درحالیکه افزودن کازئینات سدیم منجر به کاهش پارامتر مورد بررسی گردید. بنابراین، کازئینات سدیم، پروتئین نخود و روغن ذرت مکمل های خوبی برای کاهش اثرات منفی همدیگر بر خواص پس دهی روغن، گسترش پذیری و کار لازم برای برش پنیر پروسس غنی شده هستند و استفاده همزمان این سه پارامتر در پنیر پروسس در نهایت باعث بهبود ویژگی های ذکر شده آن درمقایسه با نمونه شاهد شد. نتایج این مطالعه برای توسعه پنیر و محصولات لبنی فراسودمند حاوی کازئینات سدیم، پروتئین نخود و روغن ذرت مطلوب است.