Journal of Agricultural Sciences (Tarim Bilimleri Dergisi) 2023, 29 (4): 914-923 DOI: 10.15832/ankutbd.1178258

Journal of Agricultural Sciences
(Tarim Bilimleri Dergisi)

J Agr Sci-Tarim Bili
e-ISSN: 2148-9297

jas.ankara.edu.tr

JAS

Determination of Rheological and Chemical Properties of Hemp, Rosehip Seed and Safflower Flours

Ali CİNGÖZ[a*], Nazlı ŞAHİN[b]

[a]*Department of Food Engineering, Faculty of Engineering, Tokat Gaziosmanpasa University, Tokat, Turkey*
[b]*Department of Food Engineering, Faculty of Engineering, Karamanoğlu Mehmetbey University, Karaman, Turkey*

ARTICLE INFO
Research Article
Corresponding Author: Ali CİNGÖZ, E-mail: ali.cingoz@gop.edu.tr
Received: 21 Sept 2022 / Revised: 21 Feb 2023 / Accepted: 21 Feb 2023 / Online: 24 Oct 2023

Cite this article

CİNGÖZ A, ŞAHİN N (2023). Determination of Rheological and Chemical Properties of Hemp, Rosehip Seed and Safflower Flours. *Journal of Agricultural Sciences (Tarim Bilimleri Dergisi),* 29(4):914-923. DOI: 10.15832/ankutbd.1178258

ABSTRACT

The use of alternative flour in the production of cereal products such as bread can improve the functional and nutritional properties of bread. However, the addition of substitute flour to wheat flour may have some technological, sensory, and rheological disadvantages. To eliminate these problems, alternative flours should be used in different proportions. The aim of this study is to evaluate the remaining proportions of hemp, safflower and rosehip seeds after oil extraction and to determine their chemical properties (protein, fat, ash) and to investigate the effect on rheological dough properties with Mixolab when used as a substitute for wheat flour. Accordingly, safflower, hemp and rosehip seed flour were added to wheat flour in five different amounts (5, 7.5, 10, 15 and 20%), focusing on displacement. The addition of flour increased the protein, fat, ash, phenolic content, and antioxidant capacity. It was found that the addition of 10% safflower and hemp seed flour and 7.5% rosehip seed flour had a positive effect on rheological properties. For all three flour additions, the dough development time, stability, and percent protein softening values increased, while water holding capacity decreased. Flours added at 15% or more began to negatively affect the rheological properties. This study shows that 3 different flours, which can be added up to 10%, improve the rheological properties, chemically enrich them and improve their functional properties.

Keywords: Alternative flour, Cannabis, Mixolab, Protein weakening

1. Introduction

Wheat is one of the cereals used for bread making. However, breads made from wheat flour dough are considered nutritionally inadequate (Sabanis & Tzia 2009). The partial replacement of wheat flour with non-wheat flour increases the nutritional quality and flavour of bakery products. In this context, blended flours based on wheat and other grains and non-cereal seeds (e.g., sunflower, amaranth, quinoa, lupin, chickpea, flaxseed, chia, hemp, teff) have become popular in baking technology and play several roles, such as improving the rheological properties of the dough and increasing the overall bread quality and nutritional value (Švec & Hrušková 2015). Alternative products are added to wheat flour to improve the properties of various cereal products. The use of small millet flour to improve starch digestibility in bread making (Sharma & Gujral 2019), the use of tomato seed powder to improve dough rheology (Mironeasa & Codină 2019), the production of cookies with barley flour (Jukić et al. 2022), making cakes with chickpea and chestnut flour (Gallego et al. 2022), a flour blend enriched with lentil flour (Bouhlal et al. 2019), making cakes with pigeon pea and sweet potato (Olatunde et al. 2019). However, the rapidly growing global population, the uncertainties of war, changing consumer preferences and the resulting increase in demand for food, and the growing food problem due to drought have accelerated the search for alternatives. Moreover, many projects are being carried out around the world about innovative ways to recycle waste, and the importance of zero waste is a growing concern. In line with this, this study evaluates the waste components of safflower, hemp and rosehip seeds, especially after oil extraction, in the production of cereal-based products.

Safflower (*Carthamus tinctorius* L.) is a member of the family Compositae or Asteraceae. It is an annual herbaceous plant, cultivated mainly for its seed which is used for the production of edible oil and bird seed (Malakian et al. 2011). Safflower contains pigments (cardamom), lignans, polysaccharides, essential oils and fatty acids (arachidonic acid, linoleic acid, linolenic acid, palmitic acid and stearic acid). The nutritional value is associated with a high content of proteins, essential and balanced amino acid contents (Polushkin 2007). The use of its seeds is an intriguing and promising area for the processing branches of the food industry. Although studies on safflower have mainly focused on safflower oil, there are studies on the production of healthy drinks with high antioxidant activity (Kim et al. 2002), breads enriched with functional components (Kutsenkova et al. 2019), and cookies with increased protein content (Kutsenkova et al. 2020) using safflower seeds.

Cannabis sativa L., commonly known as hemp, is an annual herbaceous plant in the Cannabaceae family that is cultivated in China and Canada (Jian et al. 2018). Industrial hemp seeds are a novel variety of cannabis which contain low levels of δ-9-tetrahydrocannabinol, with a concentration of less than 0.3%; industrial hemp seeds are now legally in dozens of countries worldwide (Wei et al. 2021). In recent years, hemp cultivation is permitted in Turkey with the condition of being controlled by the government.

Hemp has been an important source of raw materials for the processing industry, including the use of hemp seeds, fiber, clothing, chemicals, bioenergy, flour, and oil (Grof 2018). Hemp seeds contain 32% protein, 43.7% fat, and 10.3% carbohydrates, as well as a significant amount of dietary fiber, vitamins, and minerals (Bartkiene et al. 2016). The oil from hemp seeds is rich in polyunsaturated fatty acids, proteins, and essential amino acids (Malomo et al. 2014). Moreover, in the last decade, studies on hemp proteins (Mikulec et al. 2019), whose biological value is comparable to that of chicken egg white, have shown that hemp proteins have benefits such as the regulation of cholesterol and serum glucose levels with antioxidant, and anticarcinogen activity (Wei et al. 2021). Due to their high protein, dietary fiber, and oil content, hemp seeds (*Cannabis sativa* L.) have emerged as an alternative product to fortify cereal products by adding hemp flour (Bartkiene et al. 2016; Mikulec et al. 2019) to sourdough (Nionelli et al. 2018), pasta (Schettino et al. 2019), crackers (Radočaj et al. 2014), gluten-free cookies, gluten-free bread (Korus et al. 2017), and energy bars (Norajit et al. 2011).

Rose hips (Rosa spp.) are members of the genus Rosa up to 200 species are cultivated worldwide, of which 25% are found in Turkey (Murathan et al. 2016a). Rosehip fruits are rich sources of bioactive compounds, phytonutrients, and minerals (Murathan et al. 2016b). Due to these compounds, their fruits and seeds have been used for their prophylactic and therapeutic effects against infectious and inflammatory diseases, diabetes, gastrointestinal diseases, colds, diarrhea, and urinary tract diseases (Ilyasoğlu 2014). A daily consumption of 45 g of rosehip powder lowers serum levels of C-reactive protein and ceratin in patients with osteoarthritis (Szentmihályi et al. 2002). Rosehip fruits are generally consumed in dried form as nectar and fruit tea, or after processing into jam, marmalade or gel (Gül & Şen 2017). The inside of the fruit is hairy and has 20-44% seeds (Yıldız & Nergiz 1996). These seeds, by-products of the rosehip industry are generally used as animal feed, containing even greater amounts of specific nutritionally valuable and biologically active components. The seed contains 8.72% crude protein, 31.56-44.05% crude fiber, 7.97% crude oil, and 1.87% ash (Esenbuga et al. 2011). Studies on rose hips generally refer to the fruit itself and the oil extracted from the seed. Bread (Gül & Şen 2017), Turkish noodles (Koca et al. 2018) and probiotic yogurt (Gürbüz 2021) are produced from rosehip seed flour, and there remain few studies in this area.

The aim of this study is to evaluate the changes in rheological properties due to the addition of 5 different amounts of alternative flours to wheat flour using the Mixolab® instrument (gluten, kneading index, flour water removal, amylase activity and retrogradation). No study was found in which the rheological properties of hemp seed flour, rosehip seed meal, and safflower seed meal were determined using the Mixolab® instrument. In this study, the chemical properties of the alternative flours were determined and the changes in rheological properties when 3 different flours were added to wheat flour were investigated on the basis of displacement.

2. Material and Methods

2. 1. Materials

Wheat flour (0.7< % ash ≤0.8) was obtained from Birsan Birlik Gıda San. A.Ş. in Tokat, Turkey, and stored at 4 °C. Hemp seed, rosehip seed and safflower flours were purchased from Hempium Gıda in Amasya, Turkey and stored at 4 °C. All samples were degreased by the cold-pressing method. The parts remaining after oil extraction were used by grinding. All flours were stored in a cool and dry environment until use. Folin-Ciocalteu reagent was obtained from Merck (Germany), 3,4,5-trihydroxybenzoic acid (gallic acid) were obtained from Alfa Aesar (Germany), and the Ankom XT4 cartridges were purchased from Ankom Technology (USA). The other chemicals used were standard analytical standard.

2.1. Analytical methods

The dry matter and ash content of the flours were determined by the gravimetric method (AOAC 2000). The total carbohydrate content of the samples was determined by the phenol-sulfuric acid method (Geater & Fehr 2000). The micro-kjeldahl (Gerhardt KB8, Königswinter, Germany) method was used to analyse the nitrogen content of the samples (AOAC 2000) the crude protein contents of the samples were estimated using a conversion factor of 5.75. The total fat content was determined using an Ankom Fat Analyser (Ankom Technology Corp., Macedon, NY, USA), following the Ankom Technology Method.

2.3. Total phenolic content and antioxidant capacity analysis

The phenolic content of the sample was extracted by mixing 20 mL of acidified methanol (HCl/methanol/water, 1:80:10, v/v) and 1 g of sample, and the mixture was kept in a water bath (Memmert, Germany) at 25 °C for 2 hours. It was then centrifuged at 3000 rpm for 10 minutes. The clear fraction was removed and stored at -18 °C until analysis (Beta et al. 2005).

Determination of total phenolic compounds: Analysis with 2 N Folin-Ciocalteu phenol reagent was performed according to the method described by Singleton and Rossi (1965). The results were calculated as "gallic acid equivalent".

Determination of antioxidant capacity by the method FRAP: It was performed according to the method described by Benzie and Strain (1996), and the results were calculated as "trolox equivalents".

DPPH radical scavenging activity: The antioxidant capacity was determined by the DPPH (2,2 diphenyl-1-picrylhydrazil) method described by Brand-Williams et al. (1995).

2.4. Determination of rheological behaviour using mixolab®

The dough rheological investigations were performed using Mixolab® (Chopin, Tripette et Renaud, Paris, France), which simultaneously determinates dough characteristics during the process of mixing at a constant temperature, as well as during the period of constant heating and cooling. All the measurements were performed using the modified Mixolab® 'Chopin' protocol (ICC No. 173) the parameters of which are presented in Table 1. The alternative flours were added to wheat flour at 5 different rates (5, 7.5, 10, 15, and 20%) on a displacement basis. The amount of flour required for the analysis was calculated by the Mixolab® software based on the values entered for flour mixture moisture and water absorption. Values C1 at 8 min, C2, C3, C4, and C5 were used to calculate Mixolab® parameters protein weakening, breakdown, and retrogradation (Moza & Gujral 2018).

$$\text{Protein weakening (\%)} = (\text{C1 at 8 min} - \text{C2}) / \text{C2} \times 100 \quad (1)$$

$$\text{Breakdown (\%)} = (\text{C3} - \text{C4}) / \text{C3} \times 100 \quad (2)$$

$$\text{Retrogradation (\%)} = (\text{C5} - \text{C4}) / \text{C5} \times 100 \quad (3)$$

where *C1* is the torque at 8 min (Nm), *C2, C3, C4 and C5* is the torque (Nm).

Table 1- Mixolab parameters used in modified Chopin + protocol

Settings	*Values*
Mixing speed	80 rpm
Target torque (for C1)	1100 Nm
Dough weight	75.0 g
Tank temperature	30 °C
Temperature 1st step	30 °C
Duration 1st step	8 min
1st temperature gradient	15 min 4 °C / min
Temperature 2nd step	90 °C
Duration 2nd step	7 min
2nd temperature gradient	10 min -4 °C / min
Temperature 3rd step	50 °C
Duration 3rd step	5 min
Total analysis time	45 min

2.5. Statistical analysis

SPSS statistical program (SPSS, Inc., Chicago, IL, USA) was used, a variance analysis of the results (ANOVA) was performed and the differences between the groups were assessed statistically at a 95% confidence interval using the Duncan multiple comparison test.

3. Results and Discussion

Research on wheat flour substitutes continues in the food industry, which is becoming increasingly important due to global warming and drought. In this study, safflower seeds used in edible oil production and hemp and rosehip seeds used in the cosmetic industry were used. The chemical (moisture, protein, fat, total carbohydrate and ash) and (total phenolic content and total antioxidant activity) functional properties of the mixtures processed into flour were determined and are shown in Table 2.

In the analyses conducted as part of the evaluation of these valuable ingredients, which are commonly used as feed, waste and human food, hemp and safflower seed powders stand out due to their high protein content. While hemp seed powder had a protein content of 36.38% and safflower seed powder had a protein content of 35.27%, the value for rosehip seed flour was 12.93%. The pre-drying process of the solids remaining after oil extraction before milling reduced their moisture content and ranged from 4.53 to 6.96%. As the oils were removed from the products by cold pressing, about 6% oil was detected. The ash content of the flours ranged from 1.50-13.12%. Safflower seed, which stands out with an ash content of 13.12%, has high mineral and fiber properties. Kutsenkova et al. (2020) found that the protein content of the safflower seed was 17.6% and the oil content was 38.3%, while Gül and Şen (2017) in their study found the protein content of rosehip seed flour at 7.4%, the oil content at 4.6%, and the ash content at 2.3%, while Xu et al. (2021) reported the protein content of hemp seed as 24.8%, oil content as 35.5%, and ash content as 5.6%. In another study, rosehip seeds were reported to contain 31.56-44.05% crude fiber, 7.97% crude oil, and 1.87% ash (Esenbuga et al. 2011). The results of the chemical analysis obtained in our study were higher than those reported in the literature. The reason for this is the use of defatted waste products instead of direct seed flours in the study.

The results of the total phenolic content and total antioxidant capacity of the samples are shown in Table 2. The total phenolic content of all three flours was determined in gallic acid equivalents and varied between 2405-5160 µg GAE/g. Rosehip seed flour contains the highest total phenolic content of 5160 µg GAE/g. The total antioxidant capacity was determined by two different methods (DPPH and FRAP). As for total phenolic content, the highest total antioxidant capacity was found in the rosehip seed flour. The DPPH values of the hemp, safflower, and rosehip samples were 61.44, 68.12, and 71.56 µM TE/100g, respectively, and the values of FRAP were 48.32, 65.72, and 65.92 µM TE/100g, respectively. Yu et al. (2013) reported in their study that safflower seeds had a total phenolic content of 55.52 mg GAE/g.

Table 2- Chemical composition of flours

Properties	*Control flour*	*Hemp seed flour*	*Rosehip seed flour*	*Safflower flour*
Moisture (%)	13.48±0.23^{a}	4.53±0.14^{d}	6.96±0.23^{b}	5.14±0.16^{c}
Protein (g/100 g of dry weight)	13.20±0.45^{b}	36.38±1.02^{a}	12.93±0.64^{b}	35.27±0.96^{a}
Fat (g/100 g of dry weight)	0.80±0.10^{d}	5.64±0.61^{c}	6.45±0.30^{a}	6.12±0.46^{b}
T. Carbohydrate (g/100 g of dry weight)	72.23±1.30^{b}	52.13±1.06^{c}	79.12±1.23^{a}	45.49±1.28^{d}
Ash (g/100 g of dry weight)	0.75±0.14^{d}	4.85±0.08^{b}	1.50±0.06^{c}	13.12±0.71^{a}
Total phenolic content (µg GAE/g)	616.47±14.12^{d}	2405±45.78^{c}	5160±51.05^{a}	4762±38.46^{b}
Total antioxidant activity (µM TE/100g)	14.16±0.87^{d}	61.44±0.80^{c}	71.56±1.06^{a}	68.12±0.88^{b}
Total antioxidant activity (µM TE/100g)	29.60±1.32^{c}	48.32±0.48^{b}	65.92±1.08^{a}	65.72±0.75^{a}

^{a,b,c}: Values indicated by different letters in the same line are statistically significantly different ($p<0.05$)

Mixolab® (Chopin Technologies, Villeneuve la Garenne, France) is an instrument that is used to determine the rheological quality of the flour during bread making. The Mixolab technique allows the complete characterization of the flours in terms of the quality of proteins by determining their water absorption, stability, elasticity, and weakening properties; starch behavior during gelatinization and retrogradation; consistency modification when adding additives and enzymatic activity of the proteases, amylases, etc. (Stoenescu et al. 2011). This device provides, with one single test, a complex analysis of the rheological properties of wheat flour dough, considering dough behavior during mixing, protein coagulation, heating-up behavior at enzyme activity intensification, and starch gelatinization and retrogradation during the final cooling (Blandino et al. 2015).

In this study, hemp seed flour, safflower seed flour, and rosehip seed flour were added to wheat flour in five different ratios (5, 7.5, 10, 15, and 20) and the rheological properties of the obtained flour mixtures were determined using the Mixolab instrument. The rheological results obtained are shown in Table 3. Mixolab torque curves of flours from hemp seed flour, safflower powder and rosehip seed flour are shown Figure 1. As seen in Table 3 the addition of three flours increases the dough development time. The dough development time, which was 1.28 minutes for the control flour, increased to 3.33 minutes for safflower flour, 4.67 minutes for hemp seed flour, and 1.59 minutes for rosehip seed flour. The dough development time increased until the addition of 15% hemp seed flour and began to decrease at 20%. The addition of rosehip seed flour resulted in a smaller increase in dough development time compared to the other two flour additions. This decrease is thought to be caused by the rich phenolic content of rosehip seed flour (Table 2). This can be explained by the ability of the phenolic compounds to react with the sulfhydryl groups of the gluten protein or to increase the rate of protein sulfhydryl-disulfide interchanges (Welc et al. 2022). In a previous study, the addition of phenolic acids to dough was reported to decrease kneading time, tolerance, elasticity, and bread volume (Han & Koh 2011).

Table 3- Results of Mixolab analysis of hemp seed flour, safflower flour and rosehip seed flour added in different amounts

Sample	Ratio	Development time (min)	Stability time (min)	C2 Torque (Nm)	C3 Torque (Nm)	C4 Torque (Nm)	C5 Torque (Nm)	α	β	ⱴ	Water absorption (%)
Control	0.0	1.28±0.01^{h}	3.73±0.05^{n}	0.33±0.01^{d}	1.64±0.01^{cd}	1.41±0.02^{c}	2.11±0.06^{a}	-0.090	0.590	-0.016	57.50±0.06^{a}
Safflower powder ratio (%)	5.0	1.23±0.02^{hı}	6.20±0.11^{jı}	0.27±0.01^{f}	1.54±0.02^{e}	1.30±0.01^{d}	1.75±0.02^{c}	-0.082	0.078	-0.018	56.20±0.05^{b}
	7.5	1.37±0.01^{g}	6.36±0.06^{ı}	0.28±0.03^{f}	1.51±0.03^{f}	1.31±0.01^{d}	1.73±0.03^{cd}	-0.073	0.237	-0.044	56.10±0.04^{b}
	10.0	1.56±0.05^{f}	5.53±0.07^{k}	0.25±0.02^{g}	1.48±0.02^{g}	1.30±0.01^{d}	1.72±0.02^{d}	-0.074	0.106	-0.014	55.60±0.04^{b}
	15.0	3.12±0.08^{e}	5.27±0.02^{l}	0.23±0.03^{gh}	1.41±0.02^{h}	1.27±0.02^{e}	1.73±0.03^{cd}	-0.060	0.060	-0.028	56.30±0.03^{b}
	20.0	3.33±0.06^{d}	4.55±0.08^{m}	0.23±0.02^{gh}	1.34±0.03^{ı}	1.22±0.03^{ef}	1.65±0.04^{e}	-0.062	0.144	-0.060	56.00±0.05^{b}
Hemp seeds flour ratio (%)	5.0	1.53±0.02^{f}	6.88±0.14^{h}	0.34±0.05^{d}	1.60±0.02^{d}	1.40±0.01^{c}	1.74±0.01^{c}	-0.086	0.394	-0.052	57.70±0.06^{a}
	7.5	3.47±0.11^{d}	7.38±0.11^{g}	0.35±0.04^{d}	1.65±0.01^{c}	1.44±0.02^{b}	1.79±0.02^{bc}	-0.100	0.550	-0.004	56.60±0.03^{b}
	10.0	4.53±0.12^{b}	7.46±0.06^{f}	0.32±0.02^{de}	1.57±0.01^{e}	1.47±0.01^{a}	1.81±0.02^{b}	-0.090	0.297	0.014	55.75±0.04^{c}
	15.0	4.67±0.10^{a}	7.88±0.11^{d}	0.30±0.01^{e}	1.53±0.03^{ef}	1.38±0.02^{c}	1.82±0.01^{b}	-0.112	0.282	-0.004	55.20±0.11^{c}
	20.0	3.95±0.09^{c}	6.75±0.09^{h}	0.28±0.01^{f}	1.47±0.02^{g}	1.35±0.01^{d}	1.79±0.02^{bc}	-0.076	0.444	-0.024	54.50±0.13^{c}
Rosehip seed flour ratio (%)	5.0	1.53±0.02^{f}	7.53±0.03^{ef}	0.37±0.01^{c}	1.55±0.01^{e}	1.24±0.01^{e}	1.59±0.01^{f}	-0.088	0.356	-0.036	56.90±0.09^{b}
	7.5	1.44±0.01^{g}	8.28±0.10^{b}	0.38±0.02^{ab}	1.63±0.01^{cd}	1.25±0.02^{e}	1.54±0.01^{g}	-0.089	0.395	-0.067	56.55±0.10^{b}
	10.0	1.07±0.01^{j}	7.60±0.11^{e}	0.38±0.02^{ab}	1.68±0.01^{b}	1.20±0.01^{f}	1.53±0.01^{g}	-0.102	0.424	-0.063	56.10±0.08^{b}
	15.0	1.59±0.03^{f}	8.96±0.15^{a}	0.39±0.02^{ab}	1.70±0.02^{a}	1.10±0.03^{g}	1.46±0.01^{h}	-0.101	0.475	-0.054	55.80±0.08^{c}
	20.0	1.17±0.06^{hı}	8.18±0.11^{c}	0.40±0.01^{a}	1.70±0.02^{a}	0.95±0.04^{h}	1.29±0.05^{ı}	-0.106	0.492	-0.041	55.70±0.10^{c}

$^{a,b,c,\ldots ı}$: Values indicated by different letters in the same column are statistically significantly different ($p<0.05$)

As shown in Table 3, the addition of alternative flours decreases the water holding capacity. The water holding capacity, which was 57.5% for the control flour, decreased to 55.6% for safflower flour, 54.5% for hemp seed flour, and 55.7% for rosehip seed flour. The development time refers to the time elapsed until the dough first begins to form (Rosell et al. 2007). The reduction of it is technologically desirable (Pala 2012). The dough stability (min) is lower than 11% of the maximum consistency reached during the mixing. The dough stability time of the samples are shown in Table 3 and Figure 1. The stability value, which was 3.73 minutes for the control, increased to

6.37 minutes for safflower flour, 7.46 minutes for hemp seed flour, and 8.96 minutes for rosehip seed flour. Rosehip seed flour provided the highest value for dough stability. The high protein content of the added alternative flours increased the stability values.

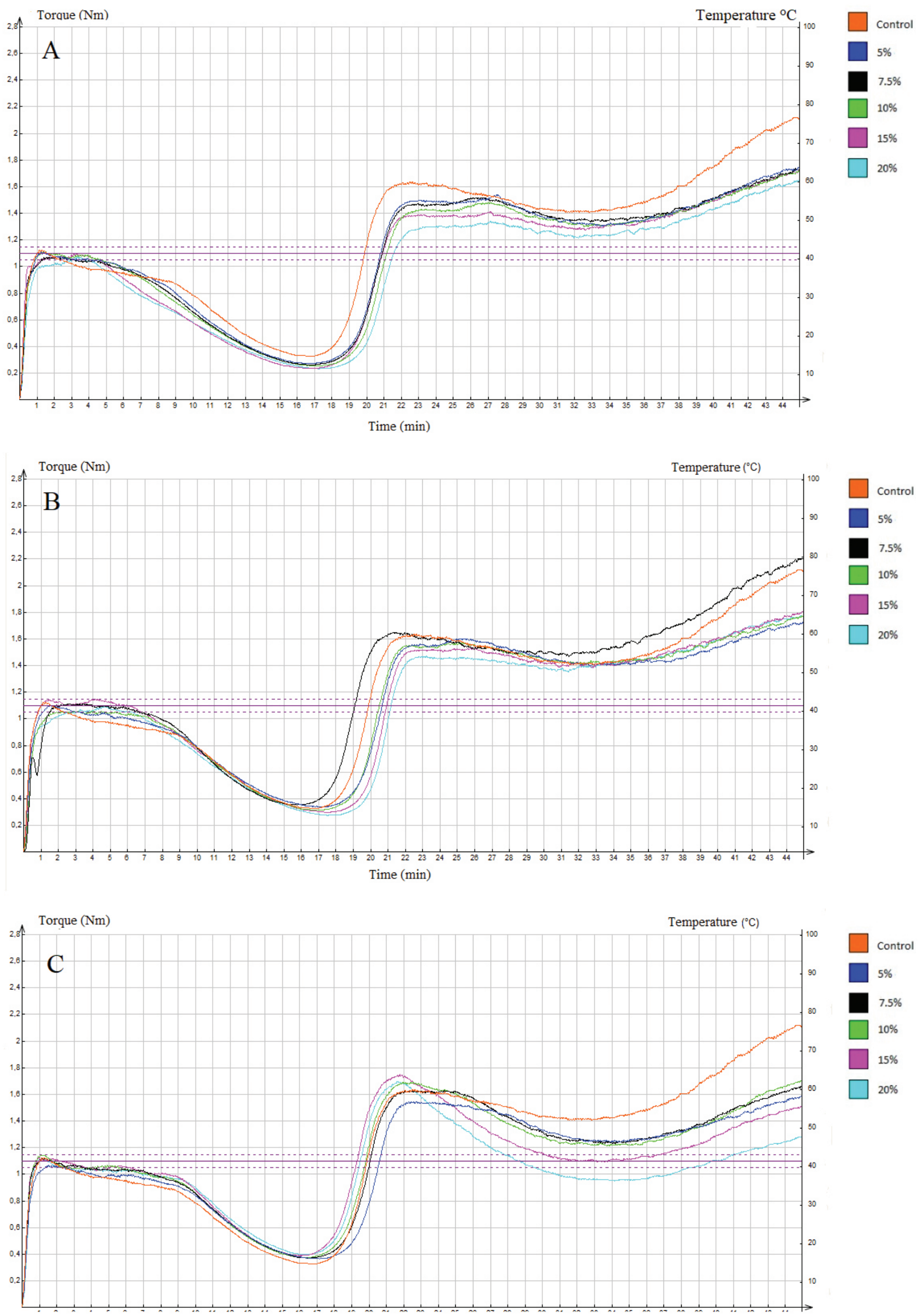

Figure 1- Mixolab torque curves of alternative flours (A) Mixolab torque curves (Nm) of safflower powder flours; (B) Mixolab torque curves (Nm) of cannabis seeds flours; (C) Mixolab torque curves (Nm) of rosehip seed flour

While the C2 torque value of 0.49 Nm in the control flour decreased with the addition of safflower and hemp seed flour, it increased with the addition of rosehip seed flour (Table 3). The rosehip seed flour addition increased the C2 value above 0.40. It is desirable that the proteins in the dough do not weakening during kneading and maintain the network structure (Cappelli et al. 2020). It is predicted that the structural properties of the proteins in rosehip flour are more robust. The C3 torque value, which gives information about the gelatinization value of the starch, decreased with the addition of safflower and hemp flour, while it increased with the addition of rosehip flour (Table 3). It is thought that the high amount of protein (~35%) in safflower and hemp flours causes this.

It was found that C4 value of 1.41 Nm in the control flour decreased for all three flour additives, with the greatest decrease for rosehip seed flour. It was found that the addition of hemp seed flour lowered the C4 torque value less than other flours and was close to the control flour for some values (Table 3). It is found that the addition of rosehip seeds and safflower flour to the dough causes the starch gel formed during cooking to break down more quickly. It is thought that the addition of hemp seed flour causes the structure of the starch gel to remain stable for a longer period of time (Figure 1).

The viscosity of the dough increased due to the retrogradation of starch as the temperature decreased from 90 °C to 50 °C. C5 value, which was 2.11 Nm for the control flour, decreased to 1.65, 1.74 and 1.29 Nm, respectively, with the addition of safflower, hemp and rosehip seed flour. The addition of safflower, hemp, and rosehip seed flours to wheat flour reduced the water-holding capacity, the mesh structure of the dough was damaged due to the proportional reduction in the amount of gluten, and staling accelerated due to the breaking of the gel structure of the starch.

It has been found that the addition of safflower flour to wheat flour increases the α angle, while the addition of hemp and rosehip seed flour decreases it. β, which was 0.590 for the control flour, decreased to 0.060 with the addition of safflower flour, to 0.282 with the addition of hemp flour, and to 0.356 with the addition of rosehip seed flour (Table 3). The addition of rosehip seed flour is noticeable as the flour that has the least effect on dough viscosity. The gamma angle increased for all three flour additives, with the increase being greatest for rosehip seed flour. It was considered that the increase in the amount of phenolic content, that could act as amylase inhibitor caused this increase. An increase in γ angle may be an indication of slowing amylase activity or a decreasing rate of enzymatic degradation. It should be taken into account that reduced amylase activity may have negative effects on the rising of the dough and the internal pore structure.

Table 4- Mixolab characteristics of hemp seed flour, safflower flour and rosehip seed flour added in different amounts

Sample	*Ratio*	*C1 (Nm)*	*Protein weakening (%)*	*Breakdown (%)*	*Retrogradation (%)*
Control	0.0	0.93±0.01[c]	64.65	14.17	33.41
Safflower powder (%)	5.0	0.89±0.01[d]	69.38	15.33	25.71
	7.5	0.88±0.02[d]	68.89	13.74	24.33
	10.0	0.82±0.03[e]	69.15	12.17	24.61
	15.0	0.74±0.02[f]	68.03	9.57	26.11
	20.0	0.71±0.01[g]	67.32	9.26	26.23
Hemp seeds flour (%)	5.0	0.94±0.03[c]	63.96	12.62	19.60
	7.5	0.99±0.05[a]	64.38	10.92	33.50
	10.0	0.96±0.02[b]	66.42	8.46	20.35
	15.0	0.96±0.02[b]	69.10	9.66	23.75
	20.0	0.92±0.03[c]	70.04	8.03	24.26
Rosehip seed flour (%)	5.0	0.94±0.02[c]	61.25	19.55	22.02
	7.5	0.99±0.01[a]	61.13	21.40	21.16
	10.0	0.97±0.02[ab]	61.05	29.36	23.59
	15.0	1.00±0.01[a]	60.01	33.21	21.51
	20.0	1.01±0.01[a]	59.76	43.74	25.53

[a,b,c,...g]: Values indicated by different letters in the same column are statistically significantly different ($p<0.05$). C1 is dough consistency after mixing; percent protein weakening is (C1at 8 min-C2)/C1%; C3 is peak consistency; percent breakdown is (C3-C4)/C3%; percent retrogradation is (C5-C4)/C5%

The Mixolab behavior of different flours is shown in Table 4. The value of C1 varied significantly within the varieties and ranged from 0.71 to 1.01 Nm for the alternative flour blends. After 8 minutes of mixing the temperature begins to increase (the viscosity of the dough decreases), excessive kneading leads to a weakening of the protein and a decrease in dough strength, and a minimum torque

C2 is reached. This decrease in dough strength is caused by physical protein breakdown and protein denaturation during heating. Mixolab measures protein weakening as a function of mechanical work (Rosell et al. 2007). The percent protein weakening (%) was calculated for the samples and ranged from 59.76 to 70.04% for alternative flour blends and 64.95% for control flours. The percent protein weakening of rosehip seed flour, which has a lower protein content than the other two flours, was found to be lower. Rosell et al. (2010) reported that the addition of dietary fiber to wheat flour may hinder protein folding and delay protein weakening. The percent breakdown (%) of flours was reported (Table 4) and ranged from 8.03 to 43.74% for alternative flour blends and 14.17% for control flour, respectively. The amylose chains recrystallize upon cooling, resulting in starch gelation (Rosell et al. 2010). The percent retrogradation of the flours was estimated and ranged from 19.60 to 33.50% for the alternative flour blends and 33.41% for the control flour, respectively. Retrogradation is predicted to decrease as a result of the decrease in water-holding capacity and gluten content.

5. Conclusions

This study investigated the changes in chemical, functional, and rheological properties of doughs caused by the addition of alternative flours of hemp, safflower, and rosehip seed waste to wheat flour. The addition of safflower, hemp and rosehip seed flours increased the content of proteins and phenolic compounds. The study showed that the addition of 3 different flours in 5 different amounts affected the rheological properties of the dough. It was found that the added flours increased the percent protein wake value and decreased the percent retrogradation value. It was also found that although the water holding capacity of the dough decreased to some extent, the dough development time and dough stability increased. The results showed that safflower, hemp and rosehip seed flours used as substitutes for wheat flour reduced the starch gelatinization rate. Safflower and hemp flour were found to have a greater effect on rheological properties, while rosehip flour had similar rheological properties to the control flour. Analyzing all the data obtained, it was found that the addition of 7.5% hemp seed flour, 5% safflower powder flour and 10% rosehip seed flour to wheat flour did not negatively affect the rheological properties of the dough. Furthermore, it was found that all added flours improved the protein content and functional properties of the control flour. This study aims to fill the gap in the literature for determining the rheological properties required for bread making by substituting safflower, hemp seed, and rosehip flours for wheat flour. In addition, the results of this study provide data on the potential of using three different flours, which are waste materials, in the food industry.

Data availability: Data are available on request due to privacy or other restrictions.

Authorship Contributions: Concept: A.C., Design: A.C., N.Ş., Data Collection or Processing: A.C., N.Ş., Analysis or Interpretation: A.C., N.Ş., Literature Search: A.C., Writing: A.C., N.Ş.

Conflict of Interest: No conflict of interest was declared by the authors.

Financial Disclosure: The authors declared that this study received no financial support.

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