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THE EFFECT OF COMPOSITION OF HYDROCOLLOIDS ON PROPERTIES OF GLUTEN-FREE BREAD*

Halina Gambuś, Marek Sikora, Rafał Ziobro

Agricultural University of Cracow

Abstract. Sensory parameters of gluten-free bread depend on the amount and type of hydrocolloids used as gluten replacers, as this determines interactions between them and starch, which is the main component of dough. The evaluation of gluten-free breads supplemented with various amounts of guar gum, pectin and xanthan, proved that bread with addition of xanthan has higher volume in comparison with pectin-guar standard. Higher amount of xanthan resulted in a decrease of bread hardness on the day of baking and after 72 hours of storage. Bread baked with equal amounts of all hydrocolloids (recipe IV) displayed best quality parameters. The amount of free amylose in crumb extract depended on the extent of starch gelatinisation, influenced by proportions of pectin, guar gum and xanthan in the mixture of hydrocolloids.

Key words: gluten-free bread, xanthan, pectin, guar gum, coeliac disease

INTRODUCTION

Coeliac disease similiarly to phenylketonuria and diabetes is a metabolic disorder, in which there occurs a strict relation between intake of certain products and severe malfunctions of the organism [Vincenzi et al. 1989, Thompson 2001]. Moreover, there is a large number of persons with minimal or no symptoms, who are predisposed to long-term complications caused by gluten intake [Mustalahti et al. 2002, Duggan 2004].

Gluten-free diet is the only efficient treatment in coeliac disease, and the results depend on the rigorous discipline in its application, in many cases in the whole period of life [Baldo and Wrigley 1984, Duggan 2004]. The food must be completely free of any gluten, so all the products from wheat, rye, barley and oat must be replaced with corn, rice, millet equivalents and various types of starch (corn, rice, potato) or appropriate mixtures [Gambuś et al. 2001, Sanchez et al. 2002]. The formulation of new, better recipes and technologies for gluten-free products is therefore a priority [Toufeili et al. 1994, McCarthy et al. 2005].

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Corresponding author – Adres do korespondencji: Prof. dr hab. inż. Halina Gambuś, Department of Carbohydrate Technology of Agricultural University of Cracow, Balicka 122, 30-149 Cracow, Poland, e-mail: rrhgambu@cyf-kr.edu.pl

Gluten may be to some extent replaced by natural or synthetic raw materials, which can significantly swell in water and form structural equivalent of gluten network in wheat dough. The most commonly used are such hydrocolloids as pectin, guar gum, arabic gum, egg albumin, galactomannans and methylcellulose. Hydrocolloids and their mixtures impact rheology of the dough as well as its baking properties and the final bread texture. Technical difficulties during gluten-free bread production (as well as gluten-free pasta) were described by many authors [Ylimaki et al. 1991, Malcolmson et al. 1993, Sanchez et al. 2002, Ahlborn et al. 2005, McCarthy et al. 2005]. Gluten free bread, based on starch, is less tasty than traditional bread and has a high staling tendency. The crumb which after baking is wet and sticking together, on the next day becomes dry, rough and crumbly. Because home-made gluten-free bread is prepared for several days, it is very important to prevent sufficient organoleptic quality during the storage [Ylimaki et al. 1991, Malcolmson et al. 1993, Gambuś et al. 2001, Sanchez et al. 2002].

Many parameters of gluten-free bread depend on the amount and type of non-starch hydrocolloids used as gluten replacers, as this determines interactions between them and starch, which is the main component of dough. There are reports on the interactions between starch and other polysaccharide hydrocolloids, such as pectin, guar gum and xanthan gum [Funami et al. 2005, Eidam et al. 1995]. In the earlier studies on the quality of gluten-free bread the synergistic action of guar gum and pectin in the mixture with corn starch was reported [Gambuś et al. 2001].

Xanthan gum is known to be compatible with many food components, such as proteins, salts, acids and thickeners: starch, carrageenan, cellulose derivatives, gelatin and alginates. Water solutions of xanthan gum are characterized by high viscosity, even at low concentrations. They also exhibit significant viscoelasticity. These features are the result of very stiff, rod-like conformation of xanthan gum in solution [Urlacher and Noble 1997]. Weak gel matrix of xanthan gums inhibits agglomeration of fat and starch retrogradation, improves structure, eating quality and appearance of food products [Food... 1990, Gimeno et al. 2004].

The aim of the current research was to evaluate the optimum proportion of hydrocolloids: guar gum, pectin and xanthan gum that could be used for the production of gluten-free bread.

MATERIAL AND METHODS

The ingredients and recipes for gluten-free bread baking are shown in Table 1. Corn starch and corn meal were obtained from Dia-Cel, Lodz, potato starch (Superior) from ZPZ Niechlow, non-starch hydrocolloids from Hortimex, oil "Kujawski" was produced by ZT "Kruszwica", instant yeast by Lesaffere, aminoacids were acquired from Waloch and S-ka, Poland.

Laboratory bread baking was performed using simple procedure, which consisted of the following stages:

– mixing of dough ingredients to obtain uniform consistency (approx. 10 min)
– putting balanced dough portions (250 g) into baking pans
– carrying out fermentation for about 40 min, at 40°C.

Table 1. Gluten free-bread recipes used in the study
Tabela 1. Receptury na chleb bezglutenowy użyte w badaniach

Ingredients Składniki g	Receptury – Recipes			
	I	II	III	IV
Potato starch – Skrobia ziemniaczana	120	120	120	120
Corn starch – Skrobia kukurydziana	432	432	432	432
Corn meal – Kaszka kukurydziana	48	48	48	48
Pectin – Pektyna	10.53	10	9	7
Guar gum – Guma guarowa	10.53	10	9	7.06
Xanthan gum – Guma ksantanowa	–	1.06	3.06	7
Yeast – Drożdże	30	30	30	30
Sugar – Cukier	36	36	36	36
Salt – Sól	10.5	10.5	10.5	10.5
Oil – Olej	18	18	18	18
L-lysine – L-lizyna	0.23	0.23	0.23	0.23
L-threonine – L-treonina	0.23	0.23	0.23	0.23
Water – Woda	630	630	630	630

The loaves were baked in oven VIVA Meteor type MD 08/6511 at 230°C for half an hour. Four loaves were obtained basing on each recipe. After cooling (1.5 hours) they were weighted in order to calculate baking loss and bread yield [Analiza... 1983]. The volume was measured in grainy material, by rape seed displacement. The loaves not selected for analysis on the day of baking were stored in packages (used in bakery for packing) at 23-24°C and 64% relative moisture content. The analyses were performed on the day of baking and after 24 and 72 hours. Sensoric assessment was conducted on the day of baking and included taste and smell, physical appearance, colour and thickness of crust as well as elasticity and porosity of crumb. Bread quality class was established basing on overall score [PN-A74108, 1996].

To study bread aging, the parameters described below were measured every day during the whole storage period.

Texture profile was obtained using texture analyser TX-XTA with XTR1 software. Two slices 30 mm thick were cut from the centre of the loaf. Each slice was compressed to 15 mm at a plunger speed of 1. Plunger diameter was 24.9 mm and height – 30.55 mm. Hardness and cohesiveness were calculated.

Apparent amylose content in water extracts of crumb [Neukom and Rutz 1981] was measured. To this end 10 g of crumb was extracted with water. In the extracts blue value was measured as an indicator of amylose retrogradation [Morrison and Laignelet 1983]. Blue value is defined as absorbance of 10 mg iodine stained starch dissolved in 100 ml of water. It is calculated from the formula:

$$Bv = A \times 10 \text{ mg/m}$$

where:

A – absorbance at 635 nm,

m – carbohydrate content, established by anthrone method.

Size exclusion chromatography of the bread extracts [Gambuś 1997] was carried out. Analysis of the obtained fractions included: total carbohydrate assessment with anthrone method [Morris 1948] – iodine-staining at 640 and 525 nm [Praznik et al. 1983] pullulan standards were used for molecular weight calibration.

All analyses were done in duplicate.

The results (except chromatography data) were statistically analysed by one-factor Anova (STAT Skierniewice). The significance of differences was calculated by Duncan's test.

RESULTS AND DISCUSSION

In Table 2 the results of the impact of hydrocolloids and recipe on baking and quality scores of gluten-free bread are collected. Recipe I, containing equal amounts of guar gum and pectin was used as standard, because such a composition proved to be optimum in the system containing two hydrocolloids and starch [Gambuś et al. 2001].

Table 2. Baking characteristics and quality of gluten-free bread
Tabela 2. Parametry wypiekowe i jakość chleba bezglutenowego

	I	II	III	IV
Weight after cooling, g – Masa po ochłodzeniu, g	216^{b}	215^{ab}	212^{a}	215^{ab}
Volume, cm^3 – Objętość, cm^3	550^{a}	585^{b}	685^{d}	660^{c}
Baking loss, % – Strata wypiekowa, %	14.1^{b}	14.5^{b}	145^{b}	13.5^{a}
Bread yield, % – Wydajność pieczywa, %	180^{a}	178^{a}	179^{a}	181^{a}
Crumb moisture, % – Wilgotność miękiszu, %	51.7^{d}	50.9^{c}	49.5^{a}	50.2^{b}
Organoleptic score – Ocena organoleptyczna	39^{a}	39^{a}	39^{a}	40^{a}
Quality class – Klasa jakościowa	I	I	I	I

Values followed by a common letter within the same row are not significantly different (P < 0.05).
Wartości oznaczone jednakowymi literami w tym samym rzędzie nie różnią się istotnie (P < 0,05).

All loaves with the addition of xanthan gum had higher volumes than standard. The highest volume was observed in case of recipe III, with 3 g of guar gum, but the yield and baking loss were in this case comparable with standard. The increase in the amount of xanthan gum (7 g, recipe IV) resulted in slight increase in volume in comparison to standard but the highest yield and consequently lowest baking loss. At the same time, the bread reached best organoleptic scores. Moisture of the crumb on the day of baking did not differ statistically between the recipes with xanthan gum (II, III, IV), but higher value was measured for standard.

Texture profile analysis demonstrated, that the incorporation of xanthan gum into the mixture positively impacted hardness (Fig. 1) and cohesiveness (Fig. 2) of the crumb – two parameters which are most difficult to optimize for gluten-free bread [Toufeili et al. 1994, Gambuś et al. 2001, Sanchez et al. 2002]. On the day of baking the lowest hardness was observed for crumb with the smallest addition of xanthan gum (recipe II), but after three days of storage the change was comparable to standard. Loaves with higher amounts of xanthan gum (recipe III and IV) remained less hard during the period of storage, despite of the similar moisture (Table 3). The most pronounced difference in hardness between standard and recipes with higher amounts of xanthan gum was observed after three days of storage.

The least positive impact on cohesiveness of the crumb was found for recipe III, with 3 g of xanthan gum. Although on the day of baking crumb cohesiveness was much better than in standard (similarly as for other recipes with xanthan gum; Fig. 2) but after three days of storage the loaves were most crumbly of all the samples.

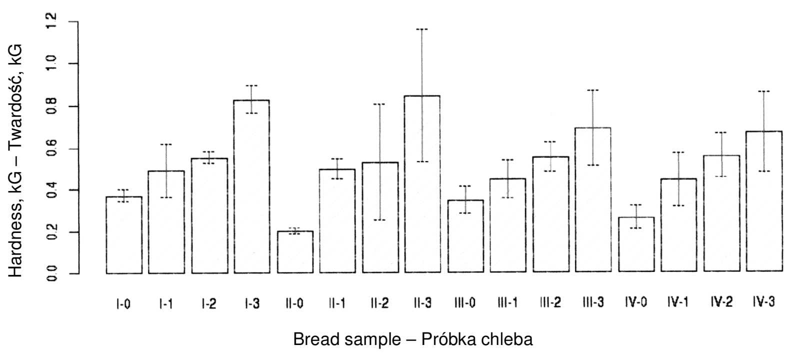

Fig. 1. Changes in crumb hardness of gluten free breads (I-IV) during storage (0-3 days)
Rys. 1. Zmiany twardości miękiszu chleba bezglutenowego (I-IV) podczas przechowywania (0-3 doby)

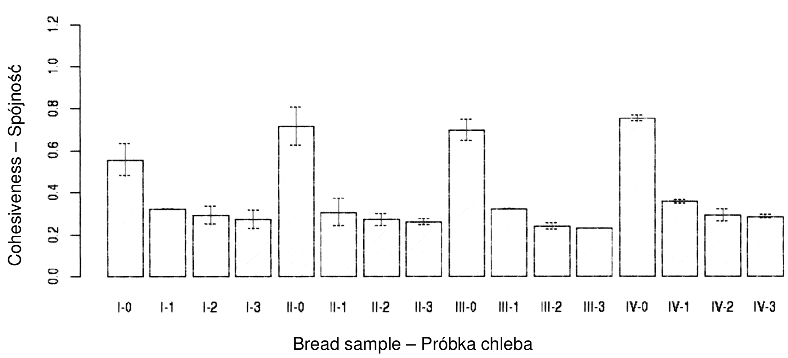

Fig. 2. Changes in crumb cohesiveness of gluten free breads (I-IV) during storage (0-3 days)
Rys. 2. Zmiany spójności miękiszu chleba bezglutenowego (I-IV) podczas przechowywania (0-3 doby)

Table 3. Changes in moisture of gluten-free bread during storage
Tabela 3. Zmiany w wilgotności pieczywa bezglutenowego podczas przechowywania

Day of storage Dzień przechowywania	Recipe Receptura	Moisture Wilgotność %
0	I	51.70[d]
	II	50.90[c]
	III	49.50[a]
	IV	50.20[b]
1	I	51.21[b]
	II	49.11[a]
	III	48.68[a]
	IV	49.33[a]
2	I	50.90[c]
	II	48.69[a]
	III	48.58[a]
	IV	49.15[b]
3	I	50.53[c]
	II	48.30[b]
	III	47.67[a]
	IV	48.35[b]

Values followed by a common letter within the same row are not significantly different ($P < 0.05$).
Wartości oznaczone jednakowymi literami w tym samym rzędzie nie różnią się istotnie ($P < 0,05$).

It seems that the introduction of xanthan gum into the mixture of hydrocolloids used as a gluten replacement significantly influenced hardness of the obtained gluten-free breads and to the lesser extent their cohesiveness.

Because the above results demonstrated the varying impact of the composition of hydrocolloids on bread quality, the following measurements were done to find out the reason of different interactions between starch and hydrocolloids used for preparation of gluten-free bread. To this end the composition of water extract of bread crumb was checked with gel permeation chromatography, and blue value was measured during the whole period of storage. The results gave the information about water soluble carbohydrates and free, unretrograded amylose in the crumb of the breads. It is well known that the maximum absorbance of amylose-iodine complex ranges from 640-660 nm, and this of amylopectin is shifted towards 520-540 nm [Polysaccharide... 1985]. The results of Praznik et al. [1983] indicate that the presence of amylose in the solution may be proved by high ratio of those values (A640/A520). In the presence of amylopectin the above mentioned ratio is low and the absolute value of A525 significantly rises. Those values should be therefore used as an indicator of starch branching in GPC fractions.

The characterisation of extract of standard bread on GPC columns is represented on Figures 3 (day of baking) and 4 (after 72 hours of storage). The presence of high amounts of soluble carbohydrates on the day of baking results from high gelatinisation of starch. The water bound by guar gum during the formation of dough was freed during

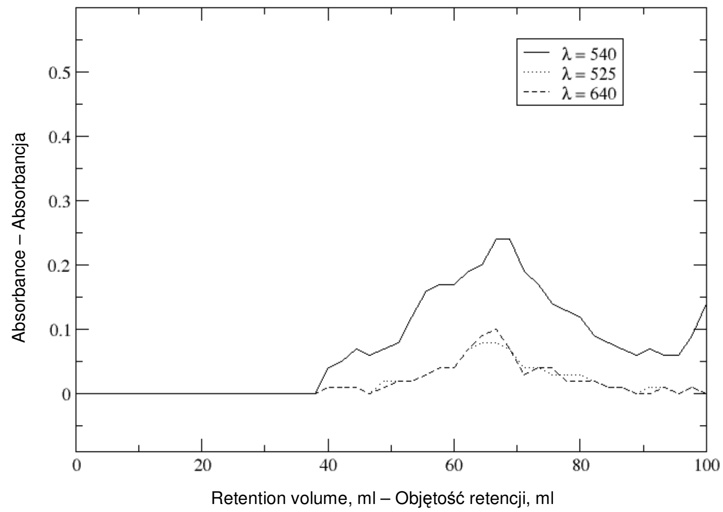

Fig. 3. Total carbohydrate content and iodine staining of crumb water extract in fractions collected from GPC columns on the day of baking of standard guar gum-pectin bread (recipe I)
Rys. 3. Zawartość węglowodanów ogółem i zdolność wiązania jodu we frakcjach wodnych ekstraktów miękiszu uzyskanych z użyciem chromatografii żelowej w dniu wypieku standardowego chleba zawierającego gumę guar i pektynę (receptura I)

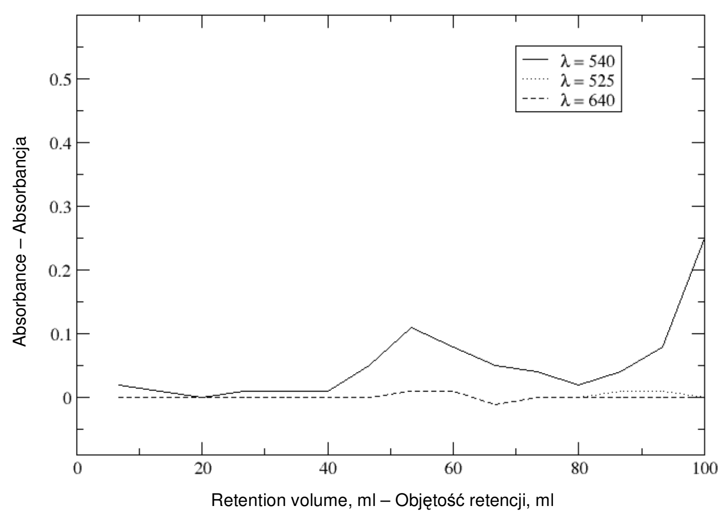

Fig. 4. Total carbohydrate content and iodine staining of crumb water extract in fractions collected from GPC columns after 72 hours of storage of standard guar gum-pectin bread (recipe I)
Rys. 4. Zawartość węglowodanów ogółem i zdolność wiązania jodu we frakcjach wodnych ekstraktów miękiszu uzyskanych z użyciem chromatografii żelowej po 72 godzinach przechowywania standardowego chleba zawierającego gumę guar i pektynę (receptura I)

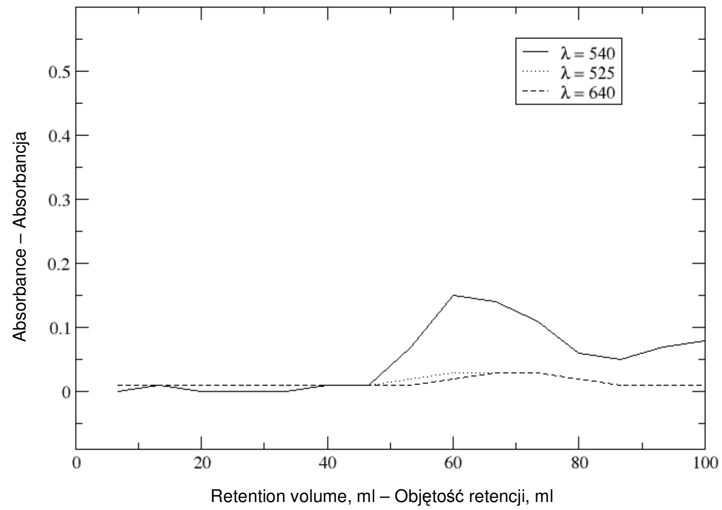

Fig. 5. Total carbohydrate content and iodine staining of crumb water extract in fractions collected from GPC columns on the day of baking of gluten-free bread (recipe II)
Rys. 5. Zawartość węglowodanów ogółem i zdolność wiązania jodu we frakcjach wodnych ekstraktów miękiszu uzyskanych z użyciem chromatografii żelowej w dniu wypieku chleba bezglutenowego (receptura II)

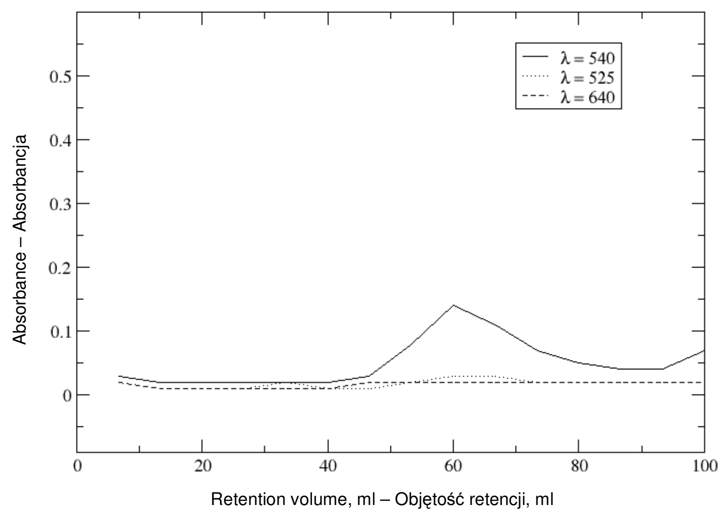

Fig. 6. Total carbohydrate content and iodine staining of crumb water extract in fractions collected from GPC columns on the day of baking of gluten-free bread (recipe II)
Rys. 6. Zawartość węglowodanów ogółem i zdolność wiązania jodu we frakcjach wodnych ekstraktów miękiszu uzyskanych z użyciem chromatografii żelowej po 72 godzinach przechowywania chleba bezglutenowego (receptura II)

baking which faciliated gelatinisation of starch granules [Michniewicz et al. 1995] and the release of initially amylose and then amylopectin. After 72 hours, the extract contained only the half of initial carbohydrates with high molecular weights, and significantly less amylose (the shift of the peak towards larger molecules; Fig. 4), which is known to retrograde several hours after baking [D'Appolonia and Morad 1981, Neukom and Rutz 1981, Ghiasi et al. 1984, Gambuś 1997].

After addition of small amounts (1 g) of xanthan gum to the mixture of hydrocolloids (recipe II) crumb extract was much lower in total carbohydrates and amylose in comparison to standard (Fig. 5). Because, as it was already mentioned, the hardness of this bread on the day of baking was the lowest of all samples, it seems, that such values were not the result of amylose retrogradation. It might be that amylose that leached during baking was bound by xanthan gum present in the dough [Christianson 1982, Katzbauer 1998]. The amounts of xanthan gum were too low for efficient self-complexation of this hydrocolloid, thus amylose-xanthan gum cross-links were favoured [Farkas 1974]. After three days of storage, the total content of carbohydrates diminished. According to the theory of bread staling this is the consequence of stepwise retrogradation of amylose and amylopectin during bread storage [Gambuś 1997].

The extract prepared from bread baked after the recipe III (Fig. 7), with 3 g of xanthan gum, contained polysachharides at the same level as in case of recipe II. The presence of amylose was however much more visible. It proves, that the gelatinisation of starch granules was not inhibited, and the leaching amylose was not complexed by xanthan gum. This could be the effect of phase separation of xanthan gum, which occurs when its molecules connect with each other. Under such circumstances the gelatinisation

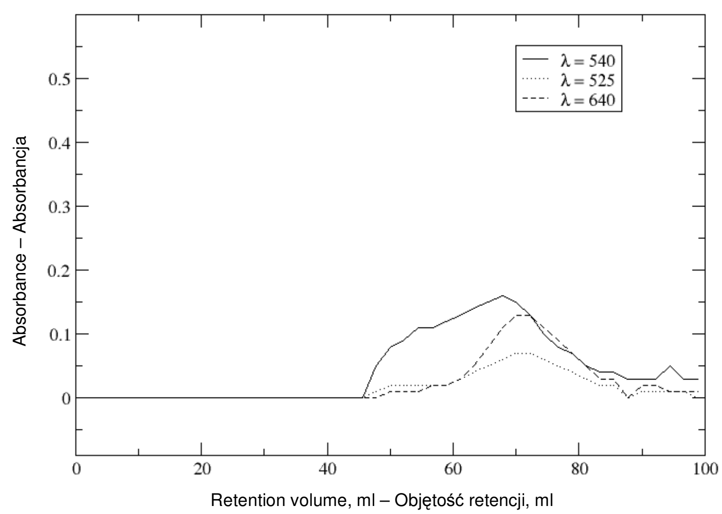

Fig. 7. Total carbohydrate content and iodine staining of crumb water extract in fractions collected from GPC columns on the day of baking of gluten-free bread (recipe III)
Rys. 7. Zawartość węglowodanów ogółem i zdolność wiązania jodu we frakcjach wodnych ekstraktów miękiszu uzyskanych z użyciem chromatografii żelowej w dniu wypieku chleba bezglutenowego (receptura III)

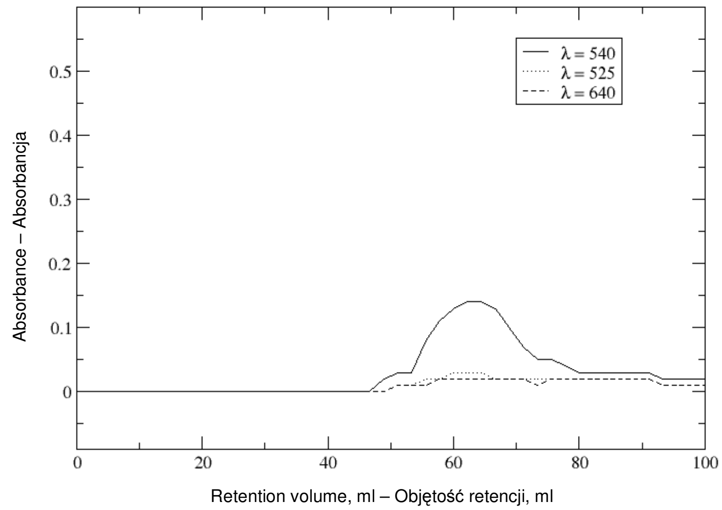

Fig. 8. Total carbohydrate content and iodine staining of crumb water extract in fractions collected from GPC columns on the day of baking of gluten-free bread (recipe III)
Rys. 8. Zawartość węglowodanów ogółem i zdolność wiązania jodu we frakcjach wodnych ekstraktów miękiszu uzyskanych z użyciem chromatografii żelowej po 72 godzinach przechowywania chleba bezglutenowego (receptura III)

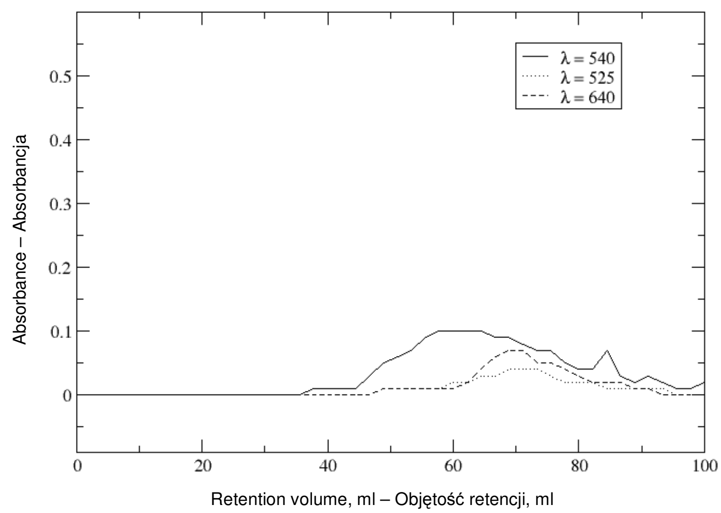

Fig. 9. Total carbohydrate content and iodine staining of crumb water extract in fractions collected from GPC columns on the day of baking of gluten-free bread (recipe IV)
Rys. 9. Zawartość węglowodanów ogółem i zdolność wiązania jodu we frakcjach wodnych ekstraktów miękiszu uzyskanych z użyciem chromatografii żelowej w dniu wypieku chleba bezglutenowego (receptura IV)

may even be enhanced [Funami et al. 2005], which would explain the presence of linear and short branched starch molecules in the extract. After 72 hours the retrogradation of both of these polymers lead to a dramatic decrease of these components.

The decrease of starch gelatinisation was found in the crumb of bread baked according to the recipe IV, with equal amounts of hydrocolloids (Fig. 9). On the day of baking, the level of carbohydrates in crumb extract was comparable with those, obtained for other recipes after 72 hours (Fig. 10). Relatively high amounts of amylose in extract indicate, that the gelatinisation process was retarded and the amylose that leached from the starch granules did not retrograde in a considerable part. It is possible that the composition of hydrocolloids favoured the interactions between molecules of different chemical structure. Such interactions might stabilize the whole system, as was observed by Eidam et al. [1995] which would be manifested in the highest volume of the bread (Table 2). The retardation of gelatinisation positively influenced hardening of bread crumb, and thus extended its shelf-life [Gambuś 1997]. During three days of storage the retrogradation of starch components was however visible, and led to the lowest values of total carbohydrates in crumb extract (Fig. 10).

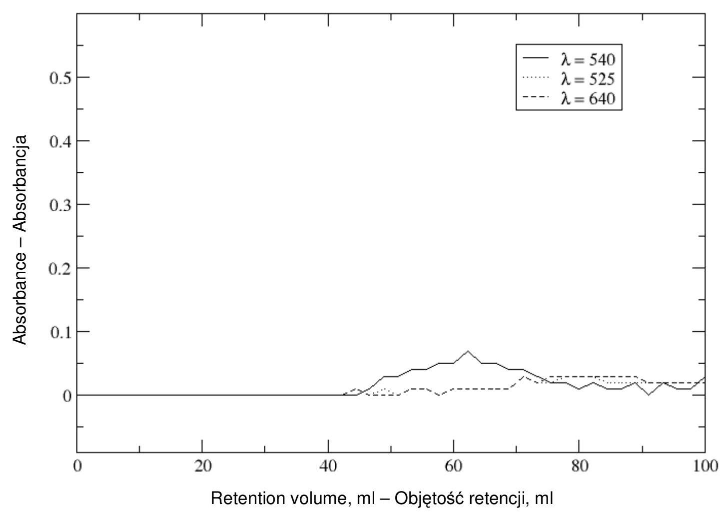

Fig. 10. Total carbohydrate content and iodine staining of crumb water extract in fractions collected from GPC columns on the day of baking of gluten-free bread (recipe IV)
Rys. 10. Zawartość węglowodanów ogółem i zdolność wiązania jodu we frakcjach wodnych ekstraktów miękiszu uzyskanych z użyciem chromatografii żelowej po 72 godzinach przechowywania chleba bezglutenowego (receptura IV)

The above mentioned changes in the level of free amylose, influenced by the proportion between applied hydrocolloids were confirmed by blue values (Fig. 11). The rate of amylose retrogradation was similar in all cases, so the changes in crumb were mainly determined by the extent of starch gelatinisation.

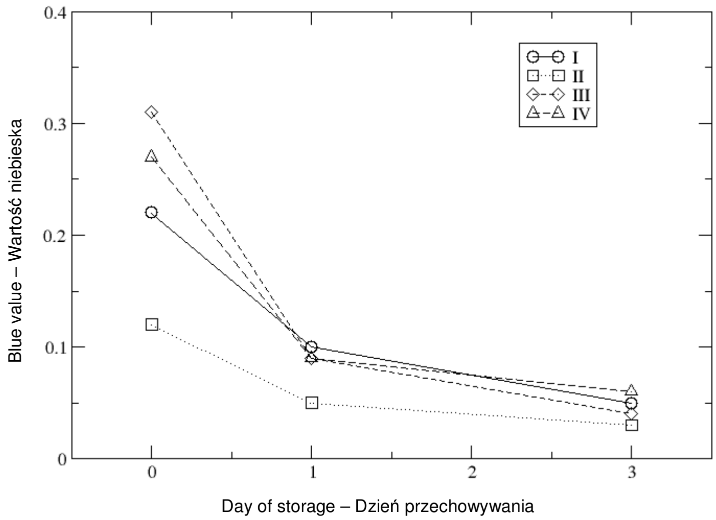

Fig. 11. Amylose retrogradation in crumb of gluten free breads, during storage
Rys. 11. Retrogradacja amylozy w miękiszu chlebów bezglutenowych podczas przechowywania

CONCLUSIONS

1. All loaves with xanthan gum displayed better volume in comparison to standard.
2. Irrespective of the share of xanthan gum, its addition to the dough led to better cohesiveness of bread on the day of baking.
3. Higher amounts of xanthan gum in mixture of hydrocolloids (3 g, 7 g) decreased bread hardness on the day of baking and after 72 hours of storage.
4. The amount of free amylose in crumb extract depended on the extent of starch gelatinisation, and not on the rate of amylose retrogradation, which was comparable.
5. The degree of starch gelatinisation was influenced by proportions and interactions of pectin, guar gum and xanthan gum in the mixture of non-starch hydrocolloids, used as gluten replacers.

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WPŁYW SKŁADU HYDROKOLOIDÓW NA WŁAŚCIWOŚCI CHLEBA BEZGLUTENOWEGO

Streszczenie. Parametry sensoryczne chleba bezglutenowego zależą od rodzaju i ilości hydrokoloidów użytych jako zamienniki glutenu, gdyż decydują one o oddziaływaniach pomiędzy nimi a skrobią, która jest głównym składnikiem ciasta. Badania na chlebach bezglutenowych, wzbogaconych o różne ilości gumy guar, pektyny i ksantanu dowodzą, że dodatek gumy ksantanowej wpłynął na większą objętość w porównaniu ze standardem pektynowo-guarowym. Większy dodatek ksantanu wpłynął na ograniczenie twardości chleba w dniu wypieku i po 72 godzinach przechowywania. Chleb wypieczony z równymi ilościami wszystkich hydrokoloidów (receptura IV) wykazywał najlepsze parametry jakościowe. Ilość wolnej amylozy w ekstraktach miękiszu zależała od stopnia skleikowania skrobi, na co miały wpływ proporcje pomiędzy pektyną, gumą guar oraz ksantanem w mieszaninie hydrokoloidów.

Słowa kluczowe: chleb bezglutenowy, ksantan, pektyna, guar, celiakia

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