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THE INFLUENCE OF REDUCING AND OXIDISING AGENTS ON RHEOLOGY OF WHEAT FLOUR DOUGH

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Abstract

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The effects of a reducing agent (L-cysteine hydrochloride monohydrate), oxidising agents (inactivated dry yeast, L-tyrosine) and other two amino acids (L-threonine and L-tryptophan) on the rheological characteristics of wheat flour dough and its quality were studied. The addition of oxidising agents increases the values such as tenacity and decreases extensibility which were measured on the alveograph. From the consistograph values, weakening of the dough also decreases. While reducing agents increase the extensibility and weakening of the dough, they decrease its deformation energy. The addition of L-tryptophan caused a higher increase in tenacity of the dough but on the other hand a larger decrease in extensibility and deformation energy. L-tryptophan caused better stability of the dough. L-treonine was found to be an oxidising agent. The dough with L-threonine weakened rapidly and therefore fast preparation of the dough is necessary. It means that the time of mixing and dough proofing must be reduced.

additive, dough, gluten, amino acid, rheology

Wheat classes are based on hardness, colour and growing season of wheat varieties. Hard wheat has a higher protein content than soft wheat and is used mostly for yeast-raised products. Soft wheat is used mostly for chemically leavened products.

Viscoelastic properties of a gluten protein matrix have a fundamental impact on the rheological properties of wheat bread dough. When the dough is developed by mixing, gluten proteins form a continuous three-dimensional viscoelastic network throughout the dough embedding discrete rigid filler, i.e. starch granules. This quasi-solid structure is infiltrated into aqueous phase composed of soluble polysaccharides, proteins and emulsified lipids, called dough liquor, and closes dispersed gas phase in form of air bubbles. Therefore, wheat gluten can be considered a simplified model of a viscoelastic skeleton of dough. Essentially, wheat gluten is composed of polymerised glutenins and gliadins, which have recently been referred to as wheat prolamins (Shewry *et al.*, 1994; Shewry and Tatham, 1997). The three-dimensional structure of gluten matrix is stabilised by covalent (disulfide) bonds, hydrogen and non-covalent ionic bonds and hydrophobic interactions (Popineau *et al.*, 1994; Shewry *et al.*, 1994; Shewry and Tatham, 1997; Belton, 1999; Lindsay and Skerrit, 1999). Recognition and understanding of the relationship between viscoelastic properties of gluten and both density and distribution of cross-links forming the three-dimensional gluten structure still remain the main objectives of cereal biochemistry and rheology (Bloksma, 1990; Cornec *et al.*, 1994; Shewry *et al.*, 1994; Janssen *et al.*, 1996; Shewry and Tatham, 1997; Belton, 1999; Lindsay and Skerrit, 1999; Lefebvre *et al.*, 2003).

Additives bring an improvement in the machine ability of dough as well as the quality and other sensory attributes of the finished product. The use of additives helps not only improve but also maintain the quality of bakery products. Some additives are used by the milling industry and others are used by the bakery industry. Additives can be divided into oxidants and reductants.

Cole (1973) reported that oxidants promote formation of disulphide bonds among the extended gluten molecules and thus impart gas-retaining and dough-strengthening properties to gluten films.

Moss (1974) reported that oxidising agents make the dough stiffer as a result of cross-linking of protein molecules, whereas reducing agents weaken the dough as a result of splitting of disulfide bonds, resulting in a reduction in molecular size.

It is also reported that the main reaction of oxidants is oxidation of a free sulphide group into disulphide moieties; gluten proteins of flour are thought to be the main target of this oxidation and the desirable effects of oxidants are marked strengthening or an increase in dough elasticity, improved machine ability and an increased volume of the final loaf (Stauffer, 1990b).

Reducing agents (like L-cysteine or gluthathione) belong to dough conditioner used to reduce mixing time and improve extensibility. They are added to bread to increase bakery throughput and adjust for flour variation, and in a number of other yeast and chemically leavened applications. Disulfide bonds in gluten are broken mechanically during mixing but they can be broken chemically by cysteine or glutathione reactions.

Cysteine is a protein amino acid that has a sulfhydryl group at the end of the molecule. Cysteine is important to dough reduction chemistry because it occurs in gluten protein from flour, in tripeptide glutathione from yeast, and in a free amino acid form as a synthetic reducing agent.

During mixing, gluten in flour is stretched and pulled. It means that it can be reformed during proofing and baking to provide the strength and structure needed.

Oxidising and reducing agents can be used separately. Moreover, a reducing agent can be used with a slow oxidising agent (e.g. potassium bromate) to increase gluten breakdown early in the process and gluten reformation later in the process.

The aim of the study was to compare two amino acids such as L-threonine and L-tryptophan against three known amino acids already used in flour modification (L-cysteine hydrochloride monohydrate, inactivated dry yeast and L-tyrosine). Considering chemical structure, it was investigated whether the addition of L-threonine and L-tryptophan leads to gluten networking by uncovalent disulphide bonds and hence oxidation effects. These amino acids were added at increasing concentrations and their influence on the rheological properties of wheat flour dough was evaluated.

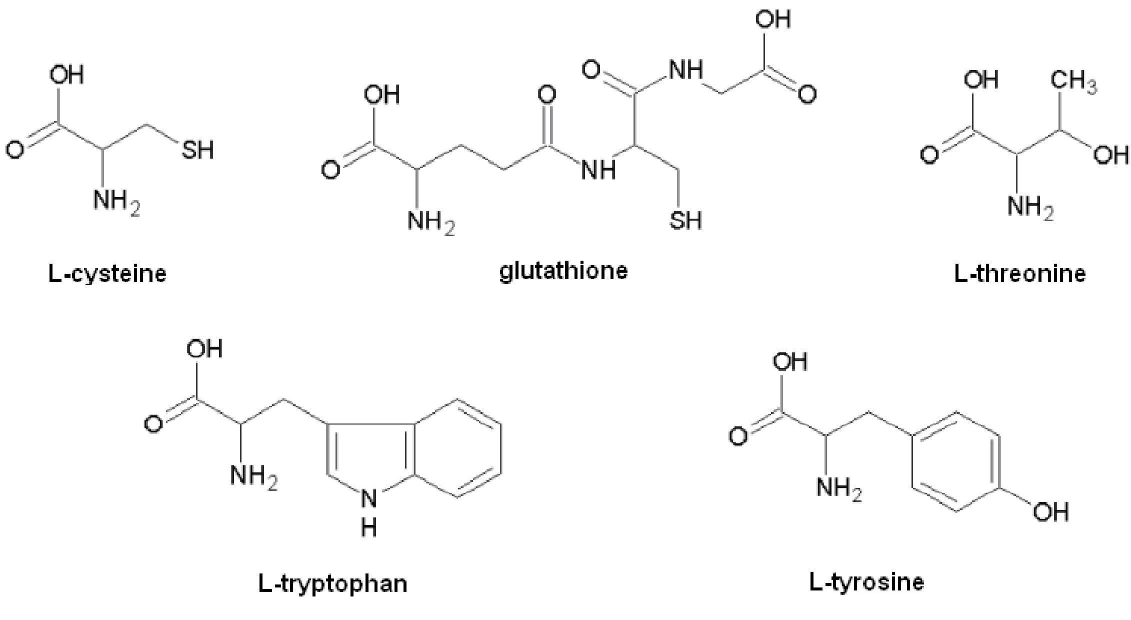
MATERIAL AND METHODS

Wheat flour

White wheat flour (moisture 14.3%, gluten in dry matter 36.4%, Falling number 339 s) provided by Penam, Kroměříž, Czech Republic, sodium chloride and redistilled water were used in the study.

Additives

Amino acids L-tyrosine, L-threonine, L-tryptophan (Merck KGaA, Darmstadt, Germany); L-cysteine hydrochloride monohydrate, inactivated dry yeast (Ireks GmbH, Eppelborn, Germany) were used for wheat flour modification within the concentration range of 0.08–0.14 wt.% (calculated on the amount of flour in dough composition). Chemical formulas are shown in Fig. 1. Totally 24 samples were prepared and analysed.



1: *Structure of aminoacids*

Rheological characteristics

The rheological characteristics of wheat flour dough were measured using Alveoconsistograph (Chopin – Tripette & Renauld, France) according to the methods AACC 54-30A and 54-50, respectively. The bread-making quality was estimated using parameters such as tenacity (P), taken as the height of the curve which measures the initial resistance to extension (forming the bubble); the length of the curve (L), which is an index of dough extensibility; the P/L ratio, which indicates the balance of the viscoelastic components of dough; and the deformation energy (W), which indicates dough strength.

Consistograph parameters evaluated were the following: maximum pressure in millibars (PrMax), TprMax as the time to reach maximum pressure in seconds, Tol (Tolerance, stability) as the time in seconds where the pressure is higher than PrMax minus 20%, drop in pressure at 250 second from PrMax minus 20% (D250) and drop in pressure at 450 second from PrMax minus 20% (D450).

RESULTS AND DISCUSSION

Figures 2 and 3 illustrate the effect of the individual oxidising or reducing agents where only the highest concentrations of the amino acids investigated are shown while Tables I and II summarize the results for all dough samples. The same concentrations of the individual amino acids were used in dough com-

I: *Alveograph characteristics of investigated doughs*

Concentration (10^{-2} %)	P (mm)	L (mm)	P/L	G (cm^3)	W (10^{-4} J)
Control dough	95	89	1.06	21.0	287
L-cysteine hydrochloride monohydrate					
1.3	53	55	0.96	16.5	74
1.5	51	52	0.98	16.0	66
1.8	46	43	1.07	14.5	53
Inactivated dry yeast					
1.3	93	93	1.01	21.4	291
1.5	92	91	1.01	21.2	280
1.8	92	93	0.99	21.4	286
3.0	93	92	1.01	21.3	288
5.0	93	91	1.02	21.3	288
10.0	107	86	1.24	20.7	315
14.0	106	91	1.16	21.2	320
L-tryptophan					
1.3	95	95	1.00	21.7	303
1.5	96	86	1.12	20.6	289
1.8	96	89	1.07	21.0	295
5.0	113	74	1.54	19.1	309
10.0	112	74	1.51	19.2	307
14.0	114	70	1.62	18.7	305
L-threonine					
1.3	97	86	1.13	20.7	294
1.5	97	82	1.19	20.2	286
1.8	98	81	1.21	20.0	282
3.0	97	79	1.23	19.8	275
L-tyrosine					
1.3	98	83	1.18	20.2	285
1.5	98	82	1.19	20.1	284
1.8	98	89	1.11	21.0	304

* *P* – tenacity, *L* – extensibility, *P/L* – configuration ratio, *G* – dough swelling, *W* – deformation energy

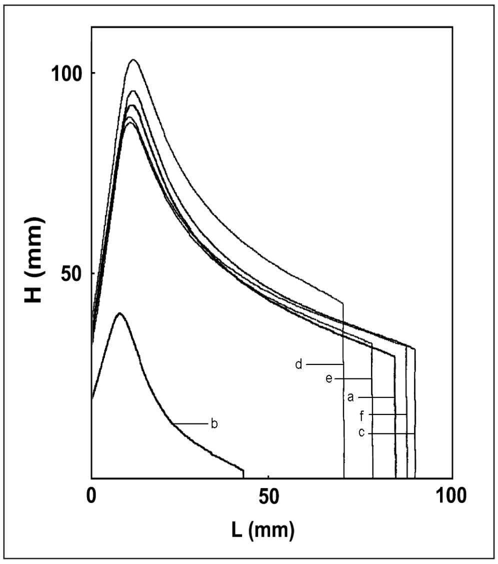
II: *Consistograph characteristics of investigated doughs*

Concentration (10^{-2} %)	PrMax (mb)	TPrMax (s)	Tol (s)	D 250 (mb)	D 450 (mb)
Control dough	2237	173	241	197	742
L-cysteine hydrochloride monohydrate					
1.3	2169	87	104	1257	1513
1.5	2150	83	95	1372	1607
1.8	2142	80	94	1405	1636
Inactivated dry yeast					
1.3	2249	164	242	204	711
1.5	2257	158	242	220	742
1.8	2157	155	294	136	638
3.0	2157	160	264	160	663
5.0	2286	160	268	188	711
10.0	2165	168	277	117	628
14.0	2218	164	278	124	652
L-tryptophan					
1.3	2240	166	279	141	690
1.5	2299	138	280	321	792
1.8	2199	164	274	129	652
5.0	2240	168	297	89	666
10.0	2254	149	304	173	693
14.0	2243	186	275	101	662
L-threonine					
1.3	2208	151	280	199	708
1.5	2281	172	266	141	708
1.8	2237	145	268	212	745
3.0	2240	158	258	220	722
L-tyrosine					
1.3	2264	142	282	170	692
1.5	2163	154	278	155	651
1.8	2206	162	261	188	719

* PrMax – maximum pressure, TPrMax – time to reach maximum pressure, Tol – time that pressure is higher than PrMAX minus 20%, D 250 – the drop in pressure at 250 seconds from PrMax minus 20%, D 450 – the drop in pressure at 450 seconds from PrMax minus 20%.
**mb (millibar)* – the device is calibrated on values mb

position. However, where the influence of amino acid on the rheological properties of dough samples was not evident at low concentrations, a higher concentration was applied.

The addition of L-cysteine hydrochloride monohydrate into wheat flour dough causes a decrease in its tenacity and extensibility compared to control dough (Table I). The higher the concentration, the larger the decrease of properties observed. However, Table II shows deterioration of dough stability and faster weakening indicated by D250 and D450 values. This effect is caused by the ability of cysteine to break disulphidic bonds and disturb the structure of gluten causing increased impalpability and stickiness of dough, which becomes less cohesive and worse in processing. Previous studies [Moss, 1974; Bloksma, 1990] showed reducing influence of cysteine on disulphidic bonds. L-cysteine hydrochloride monohydrate is a strong reducing agent and therefore only a small amount can be added to dough. It is suitable for modification of strong flours.

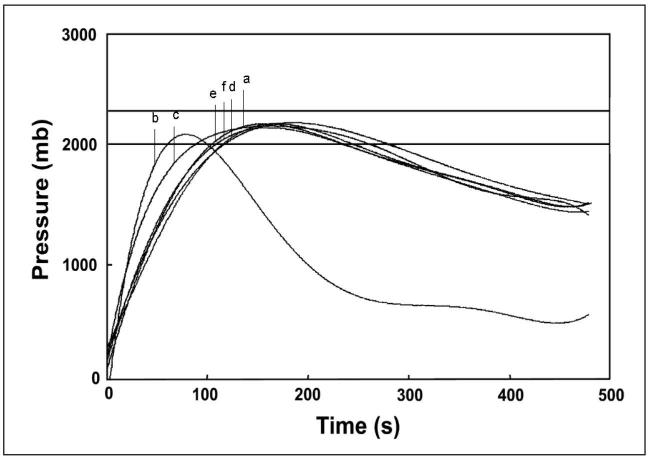


2: *Alveograph curves for the control dough (a) and dough with the highest used concentration of particular additives L-cysteine hydrochloride monohydrate (b), inactivated dry yeast (c), L-tryptophan (d), L-threonine (e) and L-tyrosine (f).*

Other three amino acids investigated, i.e. inactivated dry yeast, L-tryptophan and L-threonine, exhibited much less oxidative activity. The strongest effect is observed for L-tryptophan where, within the concentration range of 0.1–0.14 wt.%, a higher increase in tenacity and a decrease in extensibility can be seen (Table I). From the results summarised in Table II, one can notice higher stability of the dough with the addition of L-tryptophan and slower softening compared to the dough with the addition of inactivated dry yeast. Very interesting results can be seen in the modification of dough with 0.014 wt.% of L-threonine – greater strength but lower extensibility than the dough with L-tryptophan or inactivated dry yeast (Table I). At the same time, its stability is higher than that of inactivated dry yeast but lower than that of L-tryptophan (Table II). D250 and D450 stability values indicate the fastest softening of dough with L-threonine. Therefore L-tryptophan was found to be the strongest oxidative agent, followed by inactivated dry east and L-threonine. Faster softening of dough with L-threonine can be eliminated by rapid processing and reduction of mixing and fermentation times.

The last amino acid investigated (L-tyrosine) exhibits a weak oxidative effect on gluten structure. L-tyrosine is able to reinforce the gluten network and the dough becomes cohesive and more elastic. Similar information about ability of L-tyrosine to create and strengthen gluten bonds was mentioned by authors [Brady *et al.*, 1996; Peña *et al.*, 2006].

All oxidative agents were found to be suitable additives for wheat flours. Thanks to their ability to create gluten bonds, the doughs have greater strength and elasticity. Stronger oxidative additives cause a stronger dough structure and improved cohesion.



3: *Consistograph curves for the control dough (a) and dough with the highest used concentration of particular additives L-cysteine hydrochloride monohydrate (b), inactivated dry yeast (c), L-tryptophan (d), L-threonine (e) and L-tyrosine (f).*

CONCLUSION

The results showed that from the five amino acids investigated, only L-cysteine hydrochloride monohydrate is a reductant while the others are stronger or weaker oxidants. L-tryptophan was found to be the most suitable oxidising agent, followed by inactivated dry yeast and L-threonine. However, in order to eliminate fast softening of the dough with L-threonine, shorter processing time, i.e. reduced mixing and fermentation time, should be used. L-tyrosine can be used as an oxidising additive as well, but its instability in dough prevents serious practical application. Oxidants are able to strengthen the gluten network and thus increase the elasticity of dough, as a result of which bakery products have an increased volume and better porosity.

SOUHRN

Vliv redukčních a oxidačních činidel na reologii pšeničného těsta

Aminokyseliny jsou stavebními bloky bílkovin a proteinů. Mají široké rozpětí užití v potravinářských procesech a manipulacích s nutrienty, dietními doplňky, obohacovači vůně, náhražkami soli, zabarvovači, antioxidanty, konzervovadly, tvarovači, posilovači a kypřidly na těsto. Je známo, že kvalita tvorby pšeničného těsta záleží jak na kvantitě, tak na kvalitě jeho glutenových bílkovin. Gluten je viskoelastický bílkovinný komplex mající jak elastické, tak viskózní vlastnosti. Gluten obsahuje dva hlavní typy bílkovin, a to glutenin, který hlavně přispívá k elastickým vlastnostem glutenu, zatímco gliadin k viskózním vlastnostem glutenu. Tyto vlastnosti se ale právě přídavkem oxidačních či redukčních činidel dají ovlivnit, a to tak, že jednotlivé aminokyseliny reagují s glutenovou sítí, se kterou jsou schopny tvořit vazby, a tak více posilovat tuto síť (funkce oxidovadel), či naopak rozbíjet tuto glutenovou síť, jak je tomu např. u námi zmiňovaného L-cysteinu hydrochloridu monohydrátu. Tyto redukční či oxidační činidla lze samozřejmě také vhodně kombinovat, a tak docílit požadovaných viskoelastických vlastností těst.

Byly studovány účinky redukčních činidel (L-cystein hydrochlorid monohydrát), oxidačních činidel (inaktivované droždí, L-tyrosin) a dalších dvou aminokyselin L-threoninu a L-tryptofanu na reologické vlastnosti těsta a jeho kvalitu. Přídavek oxidačních činidel zvyšuje hodnoty jako je pevnost a snižoval tažnost naměřené na alveografu, a z konzistografických hodnot snižoval měknutí těsta; zatímco redukční činidla zvyšovala tažnost a měkkost těsta, snižovala deformační energii těsta. Přídavek L-tryptofanu zvýšil pevnost těsta, ale na druhé straně více snížil tažnost a deformační energii těsta. L-tryptofan dává těstu lepší stabilitu. L-threonin byl oxidačním činidlem. Těsto s L-threoninem rychleji měklo, proto rychlá příprava těsta je nezbytná. Což znamená, že čas míchání a kynutí musí být zkrácen.

přísada, těsto, gluten, aminokyselina, reologie

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