Functional Effects of Shorts in Breadmaking¹

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ABSTRACT

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Shorts, a wheat milling by-product distinct from bran and germ, has a specific, detrimental effect on loaf volume of bread. It appears that an interaction between lipoxygenase, glutathione, and a methoxyl hydroquinone type of compound is responsible for the observed effect. Evidence for the interaction of these three entities is presented. A modification of a conventional breadmaking system eliminates the detrimental effect and allows production of normal loaf-volume bread containing added shorts.

Shorts is a mixture of germ, aleurone, and pericarp layers produced on reduction rolls. Compared to bran, shorts contains more of the aleurone layer and germ. Therefore, shorts and bran may have different effects on breadmaking. Because there appear to be no reports in the literature concerning the specific effects of shorts on breadmaking, the goals of this study were to determine these effects and, if possible, overcome any that were deleterious.

MATERIALS AND METHODS

Shorts

Shorts used in this study was collected from the pilot mill of the Department of Grain Science and Industry at Kansas State University (KSU). It was milled from a mixture of hard red winter wheat varieties. Shorts as produced on the KSU pilot mill is a mixture of germ, aleurone, and pericarp layers contaminated with endosperm separated from the mill flow at four different points. A stream from the germ scalper (fraction 1), from the shorts duster (fraction 2), from the sixth midds roll (fraction 3), and from the bran and shorts duster (fraction 4).

Germ

Flaked wheat germ also was collected from the pilot mill of the Department of Grain Science and Industry at KSU. The flaked germ was ground using a Stein mill and stored at 4°C until used.

Flours

Flour A (12.2% protein $[N \times 5.7]$, 12.6% moisture, and 0.47% ash) and flour B (11.8% protein $[N \times 5.7]$, 12.4% moisture, and 0.48% ash), were provided by Ross Mills, Wichita, KS. Both flours had medium long mixing times and good loaf volume potential. Enzyme-active soy flour was obtained from Paniplus Company, Olathe, KS. Methoxyl hydroquinone (MHQ) was synthesized as described by Dakin (1909). Purified lipoxygenase was from Sigma Chemical, St. Louis, MO. All other chemicals were reagent grade.

No-Yeast Sponge Breadmaking Process

Part of the dough water was added to the flour (containing 4% nonfat dry milk, 1% enzyme-active soy flour, 6% shortening, and 0.5% sodium stearoyl lactylate) to make a no-yeast sponge. This sponge was placed in a covered fermentation bowl and allowed to stand for 2 hr. Then yeast and the rest of the ingredients were added and the dough was mixed to optimum, fermented, punched, and baked.

Pup Loaf Baking

The controls were baked by the pup loaf baking method described by Lai et al (1989). The least significant difference at the 95% confidence level for loaf volume was 20 cm³.

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Shorts fraction 4 was dispersed in water to make a 10% suspension. This suspension was centrifuged (1,500 rpm, 10 min) after 20 min of stirring. The residue was extracted with water twice, then lyophilized. The supernatants were pooled and lyophilized. This extraction scheme is outlined in Figure 1.

Defatting Fraction 4

Shorts fraction 4 was defatted with a Soxhlet using petroleum ether as the solvent (Fig. 1). Defatted fraction 4 was air dried until no solvent odor was apparent. The petroleum ether extract was dried under reduced pressure to recover the lipids.

RESULTS AND DISCUSSION

Effects of Shorts and Germ on Breadmaking

Milling data from the KSU pilot mill show that shorts contain most of the germ from the wheat kernel. Wheat contains about 3%germ by weight but only about 0.15% of the wheat is recovered as flaked germ. Thus, it appears that about 85% of the germ from wheat milled on the KSU pilot mill is recovered in the shorts. This means that the shorts (11% of the wheat) is made up of about 22% germ and 78% other material.

Mixograms showed that addition of either germ or shorts to flour reduced mixing time (Fig. 2). Addition of KIO_3 effectively prolonged the mixing time of dough containing germ. Weak et al (1977) reported that KIO_3 would overcome the effect of sulfhydrylcontaining compounds on mixing time. In addition, adding KIO_3 to the baking formula totally eliminated the detrimental effect of 3% wheat germ on loaf volume (Table I).

The addition of KIO₃ had essentially no effect on the mixing time of doughs containing shorts. KIO₃ did accelerate the breakdown of the dough as indicated by the thin tail of the mixogram curve (Fig. 2). Weak et al (1977) also reported that KIO₃ and other fast-acting oxidants accelerated the breakdown of dough. Shorts (11%) reduced loaf volume only 20 cm³ more than the dilution value. Dilution value is the reduction in loaf volume that would be expected from the dilution of gluten (Dreese and Hoseney 1982, Lai et al 1989). This indicates that the germ portion

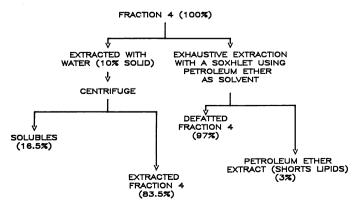


Fig. 1. Scheme for extraction of shorts.

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in shorts was not as detrimental to loaf volume as the isolated germ (Table I). Thus, there appears to be a factor in shorts that counteracts the detrimental effect of germ.

In contrast to the large effect that it had with germ, addition of KIO_3 had no effect on the loaf volume of doughs containing shorts (Table I). Higher levels of KIO_3 did not give the over-oxidized loaf characteristics commonly found with an excess of KIO_3 . These data suggest that shorts contain something that alters the effect of KIO_3 .

If an entity exists in shorts that neutralizes the effect of germ, then the addition of 3% germ plus 11% shorts should have less effect than the addition of the germ alone. If a mechanism exists that blocks the effect of added KIO₃, then adding KIO₃ to doughs containing 3% germ plus 11% shorts should not completely eliminate the effect of that added 3% germ. Germ (3%) plus 11% shorts reduced loaf volume about 34 cm³ less (90 + 20 - 76) than would be expected if there were no interaction between shorts and germ (Table I). Addition of 15 ppm KIO₃ to doughs containing 3% germ plus 11% shorts only produced 30 cm³ improvement in loaf volume (Table I). When KIO₃ was added to doughs containing 3% germ alone, loaf volume was improved by 106 cm³ (Table I). Therefore, it appears that shorts contains something that moderates the effect of KIO₃.

Simulated Shorts

A mixture of 77% bran and 23% germ was prepared to simulate shorts. Adding 11% of simulated shorts to flour reduced mixing time. Mixing time was essentially restored by addition of 15 ppm of KIO₃ to the dough (Fig. 2).

Addition of the simulated shorts to flour reduced loaf volume. Addition of 15 ppm KIO₃ to loaves containing 11% simulated shorts produced an improvement in loaf volume that would have been expected had the dough contained only the 2.5% germ portion of the simulated shorts (Table I). These data also suggest that shorts contain an entity, not found in bran, that counteracts

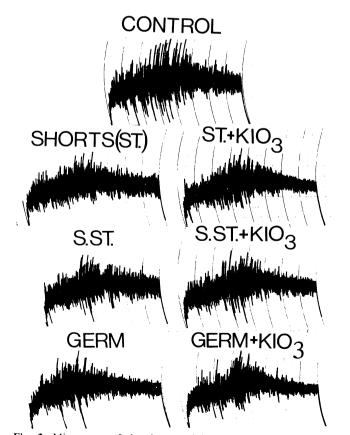


Fig. 2. Mixograms of doughs containing shorts, shorts plus KIO₃, simulated shorts (S.ST), simulated shorts plus KIO₃, germ, and germ plus KIO₃.

the effect of germ. Shorts also appear to have some component that blocks the effect of KIO₃ on dough.

Effect of Different Shorts and Shorts Fractions on Baking

The four fractions described in materials and methods (from germ scalper, from shorts duster, sixth midds roll, and bran and shorts duster) were individually collected from a mixture of newly harvested hard red winter varieties. Shorts from the newly harvested wheat ("new shorts") was much more effective in reducing loaf volume than the shorts ("old shorts") used in previous tests (Tables I and II). Both shorts samples were from Kansas hard red winter wheats (mixed varieties). The "old shorts" was from a wheat that had been stored for about one year. It is not clear if the difference in loaf volume reducing effect of those two shorts is because of wheat varieties or aging.

Baking tests showed that the four shorts fractions had different effects on loaf volume (Table II). Loaf volume and mixing time of doughs containing fractions 1 and 3 were substantially increased by KIO_3 (Fig. 3 and Table II). The added KIO_3 had little effect on the loaf volume of doughs containing fractions 2 and 4 but significantly lengthened the mixing time of dough containing fraction 2 (Fig. 3 and Table II). These data indicate that different factors in shorts are responsible for reductions in mix time and loaf volume.

Effect of Water Extraction

Fraction 4 of the shorts was extracted with water (Fig. 1). Extraction of fraction 4 with water did not reduce loaf volume (Table III). Water-extracted fraction 4 and reconstituted fraction 4 (extracted fraction 4 plus water solubles) reduced loaf volume as much as the intact fraction 4 (Table III). Apparently, the factor reducing loaf volume in shorts was not water soluble.

Addition of KIO₃ to dough containing extracted fraction 4

	•	FABLE I		
Effects of Shorts	, Germ, and	l Simulated	Shorts on	Breadmaking ^a

Treatments	Loaf Volume (cm ³)	Standard Deviation (cm ³)	V ^h
Control (flour A)	958	16.1	
11% Shorts	856	8.5	20
11% Shorts + 15 ppm KIO ₃	858	11.8	20
3% Germ	846	11.5	90
3% Germ + 15 ppm KIO ₃	952	11.9	+15
Simulated shorts			
(8.5% bran + 2.5% germ)	828	14.7	70
Simulated shorts plus KIO ₃	890	11.8	1
11% Shorts + 3% germ	798	11.5	76
11% Shorts $+ 3\%$ germ $+ KIO_3$	828	2.9	36

Numbers are averages of four observations.

^bV is the difference between the dilution value and the observed values.

TABLE II
Effect of Components of Shorts
from Newly Harvested Hard Red Winter Wheats

Treatment	Loaf Volume (cm ³)	Standard Deviation (cm ³)	Vª
Control (flour A)	965 ^b	26.5	
11% Shorts	790	13.2	92
11% Fraction 1	740	0	142
11% Fraction $1 + KIO_3^c$	850	5	32
11% Fraction 2	780	8.7	102
11% Fraction 2 + KIO ₃	805	22.9	77
11% Fraction 3	725	8.7	157
11% Fraction 3 + KIO ₃	785	7.1	97
11% Fraction 4	785	13.2	87
11% Fraction 4 + KIO ₃	806	5.8	75

^aV is the difference between the dilution value and the observed effect.

^bNumbers are averages of three observations. ^c 16 ppm KIO₃. reduced loaf volume further (Table III). Reconstitution of fraction 4 restored the KIO₃-blocking effect. Probably, the KIO₃-blocking factor is water soluble. Addition of KIO₃ to dough containing 3% germ plus 1.5% water solubles from fraction 4 of shorts resulted in an intermediate 81 cm³ improvement in loaf volume (Table III). This was still about 70 cm³ smaller than the control (Table III), indicating that the water solubles from fraction 4 contained an entity that also reacted with added KIO₃, thus, preventing the KIO₃ from fully restoring the loaf volume of doughs containing 3% germ (Tables I and III).

As shown above, the germ in shorts is not as effective as germ alone in reducing loaf volume (Table I). In addition, there appears to be a compound in shorts that reacts with KIO₃, and the reaction product reduces loaf volume. Thus, addition of KIO₃ to dough containing shorts produces a positive effect (oxidizing the germ) and a negative effect (oxidizing the compound). These two effects cancel each other and, as a result, the added KIO₃ had little or no effect on loaf volume of doughs containing shorts (Tables I and III).

Effect of Petroleum Ether Extraction

Defatted fraction 4 from shorts reduced loaf volume as much as intact fraction 4 (Table III). However, addition of KIO₃ to doughs containing defatted and nondefatted fraction 4 produced divergent results. The defatted fraction 4 gave a 50-cm³ improvement in loaf volume (807-755) as the result of KIO₃ (Table III), whereas the nondefatted fraction 4 increased only about one half that amount (775-752).

 TABLE III

 Effect of Water and Petroleum Ether Extraction

 on the Functional Properties of Shorts Fraction 4

Treatment	Loaf Volume (cm ³)	Standard Deviation (cm ³)	Vª
Control (flour A)	965 ^b	7.1	
11% Fraction 4	783	18.9	92
10.5% Water-extracted fraction 4	775	5	110
10.5% Water-extracted fraction 4 + KIO ₃	733	10.4	152
1.5% Water solubles + 3% germ	815	28	115
1.5% Water solubles + $3%$ germ + KIO ₃	896	7.5	34
11% Fraction 4	752	17.7	112
11% Reconstituted fraction 4	751	12.6	114
11% Reconstituted fraction $4 + KIO_3$	775	13.2	90
Defatted fraction 4	755	5	113
Defatted fraction $4 + KIO_3$	807	3.5	61
3% Germ + shorts lipids	887	11.5	49
3% Germ + shorts lipids + KIO ₃	918	5.8	18

^aV is the difference between the dilution value and the observed value. ^bNumbers are averages of four observations.

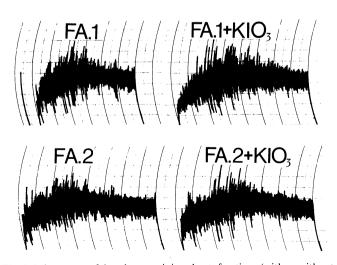


Fig. 3. Mixograms of doughs containing shorts fractions (with or without the added KIO₃).

This suggests that the compound interfering with KIO_3 action is soluble in petroleum ether. If so, addition of KIO_3 should not completely restore the loaf volume of doughs containing added germ and shorts lipids because KIO_3 should react with the interfering compound and produce a detrimental effect on loaf volume.

The addition of KIO_3 to dough containing germ plus shorts lipids failed to completely restore the loaf volume as it did to doughs containing 3% germ alone (Tables I and III). Surprisingly, shorts lipids alone substantially improved the loaf volume of doughs containing 3% germ (887 vs. 846).

That finding suggests that something in shorts lipids may interact with glutathione, and the same entity or something else may react with KIO₃ to produce a negative effect on loaf volume.

Effect of Lipoxygenase on the Loaf Volume Reducing Effect of Germ

Galliard (1986a,b) documented lipase and lipoxygenase activity in wheat bran and germ. It is possible that lipoxygenase in shorts oxidizes free fatty acids producing peroxides, which, in turn,

TABLE IV
Effects of Germ, Glutathione, and Methoxyl Hydroquinone (MHQ)
on Baking

Treatment	Loaf Volume ^a (cm ³)	Standard Deviation (cm ³)
Control (flour A) EASF ^c	960 ^b 955 ^b	20 0
EASF + 3% germ EASF + 3% germ + KIO ₃	918 ^b 957 ^b	5.8 5.8
Control (control flour B) 144 ppm Glutathione	878 653	5.8 5.8
144 ppm Glutathione + 15 ppm KIO ₃	872	16.1
30 ppm MHQ 60 ppm MHQ	900 812	11.5 5.8
30 ppm MHQ + KIO ₃ 144 ppm Glutathione + 30 ppm MHQ	663 769	9.6 21.7
132 ppm Glutathione + 30 ppm MHQ	806	13.1
132 ppm Glutathione + 30 ppm $MHQ + KIO_3$	804	18.0
EASF + mixture of 132 ppm glutathione and 30 ppm MHQ Lipoxygenase ^d + mixture of	845	17.3
132 ppm glutathione and 30 ppm MHQ	855	28.3

^aNumbers are averages of four observations.

^bNumbers are average of three observations. In these tests, 3% of the formulated flour was replaced by germ.

^cEnzyme-active soy flour (lipoxygenase) 1% of flour weight.

^d0.001% based on flour weight.

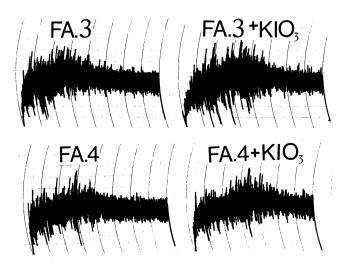


 TABLE V

 Effects of Soaking on the Activity of Shorts

Treatments	Loaf Volume ^a (cm ³)	Standard Deviation (cm ³)
Control (control B)	866	8.5
11% Shorts	839	18.9
11% Soaked shorts	913	20.2

^aNumbers are averages of four observations. Shorts or bran-shorts mixture were added to 100 g of flour.

oxidize the germ.

When we added lipoxygenase to doughs, it had no effect on loaf volume in the absence of germ. In doughs containing 3% germ, lipoxygenase substantially alleviated the detrimental effect of the germ (Table IV). Thus, lipoxygenase appeared to have an oxidizing effect similar to KIO₃.

Effects of Glutathione and MHQ

Activated, double-bond compounds such as ferulic acid bind to gluten when a thiyl radical is produced during dough mixing (Sidhu et al 1980, Danno and Hoseney 1982, Jackson and Hoseney 1986, Kivett 1987). This type of reaction has been shown to be detrimental to bread loaf volume (Moore 1984). Structurally, MHQ has a ring similar to that found in ferulic acid. The existence of MHQ in pericarp and aleurone layers was reported by Daniels (1959). Glutathione is well known to exist in germ (Sullivan et al 1936). We determined the effect of both glutathione and MHQ on baking and their response to added KIO₃.

Glutathione reduced both mixing time and loaf volume. Loaf volume could be totally restored by addition of 15 ppm KIO₃ (Table IV). Glutathione appears to mimic the effect of germ in baking. MHQ also reduced mixing time and accelerated the breakdown of dough. The effect of MHQ on loaf volume was concentration dependent. At 30 ppm (based on flour weight), MHQ improved loaf volume. At 60 ppm, MHQ slightly reduced loaf volume (Table IV). Our data also showed that MHQ interacts with glutathione and partly alleviates the detrimental effect of glutathione. When added with KIO3, MHQ was very detrimental to loaf volume even at 30 ppm (Table IV). Addition of 132 ppm glutathione plus 30 ppm MHQ reduced loaf volume about 60 cm³. Addition of KIO₃ to dough containing 132 ppm glutathione plus 30 ppm MHQ produced no effect on loaf volume (Table IV). These data indicate that a combination of MHQ and glutathione, in proper proportion, can mimic the effect of shorts on loaf volume.

MHQ would form a quinone when it is oxidized by KIO₃. The quinone could react with a thiyl radical in a manner similar to ferulic acid and reduce loaf volume (Kivett 1987). Kivett also suggested that KIO₃ oxidizes the thiols of small proteins and reduces the level of sulfhydryl-disulfide interchange. Without the interchange to relieve stress on the gluten, more disulfide bonds are broken to form thiyl radicals during mixing. Therefore, MHQ is detrimental to the bread loaf volume in the presence of KIO₃. Lipoxygenase oxidizes unsaturated fatty acids to form free radicals and peroxides, which, in turn, oxidize glutathione. The free radicals from oxidizing fatty acids react with the thiyl radicals from gluten and/or the quinones from MHQ (Sidhu et al 1980, Hoseney et al 1980, Kivett 1987). Therefore, lipoxygenase might be able to eliminate the detrimental effects of a glutathione-MHQ mixture.

To test that hypothesis, enzyme-active soy flour (0.5%, based on flour weight) or purified lipoxygenase (0.001%, based on flour weight) was added to a sponge containing mixture of 132 ppm glutathione and 30 ppm MHQ but no yeast. Yeast activity rapidly produces an anaerobic environment in fermenting dough and because lipoxygenase requires oxygen, the effects of lipoxygenase must be observed in a no yeast dough. The no-yeast sponge was allowed to stand for 2 hr before yeast was added and dough was mixed to optimum. Loaf volume was completely restored (Table IV).

Effect of Soaking on the Loaf Volume Reducing Effect of Shorts

Our finding that the detrimental effect of a glutathione-MHQ mixture (which mimics the effect of shorts on breadmaking) was eliminated by adding lipoxygenase suggested that the detrimental effect of shorts could be eliminated by soaking the shorts to allow the indigenous lipoxygenase to neutralize MHQ and glutathione. Shorts (11 g) was soaked with 13.5 cm³ water for 1 hr and then added to 100 g of flour plus other ingredients, mixed, and baked. As expected, soaking totally removed the detrimental effect of shorts (Table V).

CONCLUSION

The effect of shorts on breadmaking is very different from that of bran. An entity in shorts not present in bran can interact with germ and alleviate the detrimental effect of germ. This entity also interacts with KIO₃, and the resultant compound reduces loaf volume. Our data suggest that an MHQ-type of compound may be the interacting entity in shorts. A mixture of glutathione and MHQ in an appropriate ratio will mimic the effect of shorts on breadmaking. The detrimental effects of the glutathione-MHQ mixture can be eliminated by inclusion of lipoxygenase in a noyeast sponge. Soaking and, thus, using the indigenous lipoxygenase, totally removes the detrimental effect of shorts if breads are baked with optimum absorption.

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