

Antioxidative-like Effect of Different Cereals and Cereal Fractions in Aqueous Suspension

Pekka Lehtinen* and Simo Laakso

Laboratory of Biochemistry and Microbiology, Helsinki University of Technology, P.O. Box 6100, FIN-02015 TKK, Finland

The effect of different cereal–water suspensions on the oxidation of linoleic acid was studied. The cereals used were wheat, barley, rye, and oat. Oat suspension was the most effective inhibitor of linoleic acid oxidation and caused the dioxygenation rate to diminish by 50% at a flour concentration of 0.41% (w/v). Oat was further fractionated into fiber, soluble fiber, protein, and starch fractions. The inhibitory capabilities were highest in the soluble fiber and fiber, which caused the dioxygenation rate to diminish by 50% at a flour concentrations of 0.16% and 0.27%, respectively. The antioxidative effect appears to be due to the protection of linoleic acid from the oxidizing catalyst. The degree of inhibition correlates with the capability of the flours and flour fractions to bind linoleic acid from the aqueous system.

Keywords: *Dioxygenation; linoleic acid; lipoxygenase; inhibition; antioxidant; oat; fiber*

INTRODUCTION

Various plant materials, especially cereals and beans, contain large amounts of the lipoxygenase enzyme (EC 1.13.11.12), which catalyzes the formation of relevant conjugated diene hydroperoxides from pentadiene moieties of polyunsaturated fatty acids. In cereals, the most abundant pentadiene acids are linoleic acid, C18:2(*n*-6,9), which constitutes typically from one-third to over half of the total fatty acids, and linolenic acid, C18:3(*n*-6,9,12), the amount of which varies from 1% to 10% of total fatty acids. Plant lipoxygenases catalyze preferably the oxidation of free fatty acids, their methyl esters, and α -monoglycerides (Graveland, 1970). However, in most studies on lipoxygenases, the oxidation of di- or triglycerides or phospholipids has been shown to be very slow, except in the presence of bile salt surfactants (Piazza *et al.*, 1996; Laakso and Eskola, 1983).

The manifestation of lipoxygenase activity in cereal products during processing is governed by a multitude of factors, and the contributing factors are not fully understood. In wheat flour–water dough, the oxidation of free linoleic and linolenic acids and their monoglycerides occurs mainly within the first few minutes of dough mixing. After an initial burst, the oxidation rate diminishes and the remaining fatty acids are relatively resistant to lipoxygenase action (Mann and Morrison, 1975). The oxidized fatty acids comprise some 15–30% of total free acids and monoglycerides (Grant and Sood, 1980; Mann and Morrison, 1974, 1975). Unoxidized acids are described as bound lipids and are unextractable with hexane. The nature of binding is unknown, though inclusion in protein or starch matrix is suggested (Chung and Tshen, 1975). Not only unoxidized acids but also relevant hydroperoxides are bound by flour components (Meshehdani *et al.*, 1990). Lipoxygenases need a small initial amount of hydroperoxides for catalysis, and a reduction of the hydroperoxide below a critical level can cause a reduction in the oxidation rate (Schilstra *et al.*, 1992; Kulkarni *et al.*, 1990).

Phenolic constituents of low molecular weight present in cereals have been studied intensively as antioxidants during the recent years (Dimberg *et al.*, 1993; Tian and White, 1994). However, their low concentration in grains and the fact that their purification requires a solvent extraction have suppressed their wider use in the food processing industry. Also, the aqueous extracts from cereals have been shown to contain a heat stable inhibitor of lipoxygenase (Fretzdorff and Jördens, 1986; Nicholas *et al.*, 1981a). Nicholas *et al.* (1981a) have suggested that the inhibitor in wheat extract acts on the substrate of oxidation reaction rather than on lipoxygenase. They also partially purified the inhibitor from wheat and found that in gel filtration, the inhibitor coeluted with gliadin and that the inhibitor was inactivated after incubation with pepsin (Nicholas *et al.*, 1981b). A proteinous inhibitor of lipoxygenase is also found in blood serum and milk. In milk and serum, respectively, the inhibitor has been identified as casein (Laakso and Lilius, 1982) and albumin (Duniec and Robak, 1984). In the case of albumin the mechanism of inhibition is believed to result from binding of the substrate in a manner which renders it unavailable to the enzyme.

Thus, an antioxidative effect can arise from physical protection of the substrate. Furthermore, cereal compounds are a potential source of hydrocolloids that can act as physical antioxidants. The aims of this study were (1) to compare the suitability of major cereals and cereal products as raw material for the industrial production of antioxidatively acting compounds and (2) to test such an antioxidant in a milieu that prevails during food processing.

MATERIALS AND METHODS

Cereal Samples. Rye (cv. Voima), wheat (cv. Aura), barley (cv. Kymppi), and oat (cv. Veli) samples were harvested in 1994 and were obtained from the Agricultural Research Centre of Finland. Samples were kept at room temperature as whole grains until milled (Fritsch, Pulverisette 14, sieve ring 0.5 mm) just prior to use. Denatured flours were made by autoclaving the grains for 20 min at 120 °C, and then the grains were cooled before milling.

Oat Fractions. Oat was fractionated to protein, starch, and fiber fractions according to the wet method of Primalco

* Author to whom correspondence should be addressed (telephone +358-9-451 2561; fax +358-9-451 2564; e-mail Pekka.Lehtinen@hut.fi).

Ltd., Finland (Liukkonen *et al.*, 1992). The method consists of soaking and homogenizing of oat flour and recovering different fractions by centrifugation and sieving. Protein and starch fractions were produced at laboratory scale, whereas the fiber was fabricated industrially by Primalco Ltd. Industrially made fiber was remilled (Fritsch, Pulverisette 14, sieve ring 0.5 mm) to give a finer flour. Soluble oat fiber was made from crude fiber by extracting the fiber with 15 volumes of water for 2 h at 40 °C and lyophilization of the supernatant after centrifugation.

Chemicals. Free linoleic acid (L-1376) and soybean lipoxygenase (L-8383) were obtained from Sigma (St. Louis, MO). HPLC grade solvents were used for fatty acid analyses and extractions. Other chemicals were of reagent grade or higher grade.

Measurement of the Oxidation Rate. Oxidation of linoleic acid was followed by using polarography (Clark electrode, YSI 5300) to measure the oxygen consumption of aqueous solutions containing 0.86 mg of free linoleic acid in 0.3 mL of Tween 20/NaOH solution (Axelrod *et al.*, 1981), different amounts of milled cereal samples in 0.2 M sodium phosphate buffer, pH 7.0, and soybean lipoxygenase in 0.05 M sodium phosphate buffer, pH 8.0.

The dioxygenation activities are presented as nanokatal. One nkat corresponds to 10^{-9} mol of O_2 consumed in 1 s. The initial O_2 concentration of the buffer saturated with air was assumed to be 268 μ M. Standard deviations of multiple measurements were <10%.

Measurement of the Inhibition of the Dioxygenation Rate by Denatured Flours and Oat Fractions. A cereal sample, 0.86 mg of free linoleic acid in 0.3 mL of Tween 20/NaOH (Axelrod *et al.*, 1981) and 7.0 mL of 0.2 M sodium phosphate buffer, pH 7.0, were mixed for 100 s in a reaction cuvette. The cuvette was opened, and lipoxygenase was pipetted into the solution. The rate of oxygen consumption (roc, %/min) was recorded, and the inhibition of dioxygenation caused by the sample was calculated as follows:

$$\text{inhibition} = 100\% \times [1 - (\text{roc with the cereal sample} / \text{roc without the cereal sample})]$$

The concentration (% w/v) of the sample required to diminish the roc by 50% (i.e. inhibition = 50%) is cited as $IC_{50\%}$ concentration. The $IC_{50\%}$ concentrations are calculated from hyperbolic fitting of inhibition versus concentration of flour.

Measurement of the Lipoxygenase Activity in the Presence of the Native Flours. Cereal sample and 7.0 mL of 0.2 M sodium phosphate buffer, pH 7.0, were mixed for 100 s in a reaction cuvette. The cuvette was opened, and 0.86 mg of free linoleic acid in 0.3 mL of Tween 20/NaOH (Axelrod *et al.*, 1981) solution was added to the cuvette. The roc was recorded for 100 s and, on the basis of these values, the lipoxygenase activity inherent to the flour was calculated. Immediately after 100 s of measuring, an additional 100 or 200 μ L of soybean lipoxygenase solution (1 mg/mL) was added to the cuvette and the recording of roc was continued. The total apparent lipoxygenase activity was calculated from the roc.

Effect of Oat Flour and Oat Fiber on the Oxidation of Barley Flour Lipids in Water Suspension. Barley flour was suspended (10% w/v) in 0.3 mM $FeSO_4$. Oat flour or oat fiber, 0% or 2% (w/v), was added to the mixture. The slurry was kept at 80 °C under air, and the evaporated water was replaced with distilled water. Samples for the determination of fatty acid composition were taken daily and stored under nitrogen at -60 °C until fatty acid analysis. All experiments and every sample were done in duplicate. Standard deviations for multiple experiments were <10% and for multiple samples negligible.

Effect of Oat Fiber on the Oxidation of Heated Rapeseed Oil. To a commercial rapeseed oil (Rypsiöljy, Raisio Ltd., Finland) was added 0–5% of oat fiber. The oil was kept at 100 °C under air and mixed daily. The samples for the determination of fatty acid composition were taken weekly and stored under nitrogen at -60 °C until fatty acid analysis. All experiments and every sample were done in duplicate. Stan-

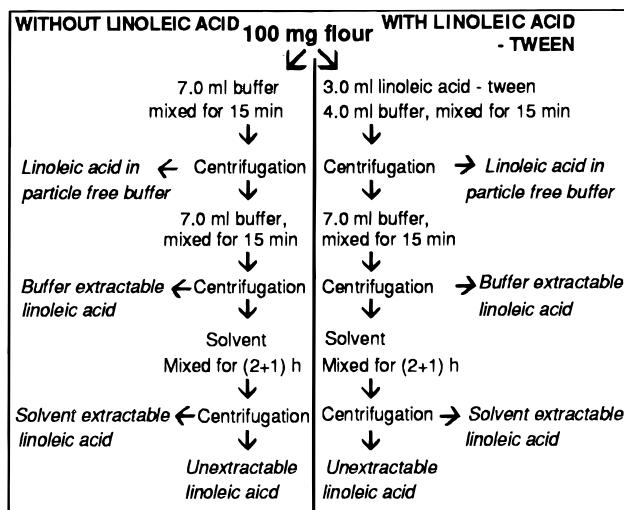


Figure 1. Schematic presentation of sample preparation.

ard deviations for multiple experiments were <5% and for multiple samples negligible.

Fate of Supplemented Linoleic Acid in Flour–Water Suspensions. Sample preparation is shown schematically in Figure 1. During the experiment the fate of extraneous linoleic acid was monitored when a surplus of the acid was added to the aqueous suspension of different native flours containing lipoxygenase activity.

To 100 mg of flour were added 8.63 mg of linolenic acid in 3 mL of Tween 20/NaOH solution (Axelrod *et al.*, 1981) and 4 mL of 0.2 M sodium phosphate buffer, pH 7.0. The tube was mixed for 15 min at 25 °C. The mixture was centrifuged at 15000g for 15 min. The supernatant (particle-free buffer) was stored under nitrogen at -60 °C until fatty acid analysis. The pellet was washed with 7.0 mL of the same buffer for a further 15 min and recentrifuged. The second supernatant (buffer extractable) was collected, and the pellet was further extracted twice (2 h + 1 h) with a total of 4 mL of chloroform/methanol (100:1) solution. Solvent (solvent extractable) and residue (unextractable) were dried under a stream of nitrogen, and fatty acids were quantified by gas chromatography. The same experiment was also done without the addition of linoleic acid. All experiments and every sample were performed in duplicate. Standard deviations for multiple experiments were <10% and for multiple samples negligible.

Analysis of Fatty Acid. Fatty acids were converted to methyl esters by saponification and methylation essentially as described by Suutari *et al.* (1990). The fatty acid contents of unextractable particle fractions were determined by adding saponification reagent straight upon the flour sample and doing the saponification and methylation similar to other samples. Absolute amounts of methyl esters were determined by adding heptadecanoic acid methyl ester as an internal standard prior to saponification. Methyl esters were identified and quantified by gas chromatography with a flame ionization detector. Methylation and gas chromatography were done in duplicate. Variation of data was negligible.

RESULTS

Apparent Dioxygenation Activity of Flour–Water Suspensions Supplemented with Lipoxygenase. The dioxygenation rate was measured in flour–water suspension containing extraneous linoleic acid and compared to the rates obtained upon supplementation of the suspension with soybean lipoxygenase. Two different amounts of lipoxygenase were added, 8.8 and 16.0 nkat.

The dioxygenation activity of flours was highest in barley (Table 1). Fretzdorff and Jördens (1986) found that the dioxygenation activity of rye was nearly twice the activity of barley when the measurements were done

Table 1. Apparent Total Dioxygenation Activities (Nanokatal) of Samples Containing 30 mg of Native or Denatured Flour and Different Amounts of Soybean Lipoxygenase

| | | no extraneous lipoxygenase | 8.8 nkat of lipoxygenase added | 16.0 nkat of lipoxygenase added |
|-----------|--------|----------------------------|--------------------------------|---------------------------------|
| native | barley | 6.4 | 7.0 | 8.9 |
| | wheat | 3.9 | 7.6 | 11.6 |
| | rye | 3.3 | 7.3 | 9.7 |
| | oat | 0.3 | 3.0 | 7.7 |
| denatured | barley | 0.0 | 6.8 | 11.7 |
| | wheat | 0.0 | 7.4 | 12.6 |
| | rye | 0.0 | 7.0 | 11.8 |
| | oat | 0.0 | 4.6 | 8.9 |

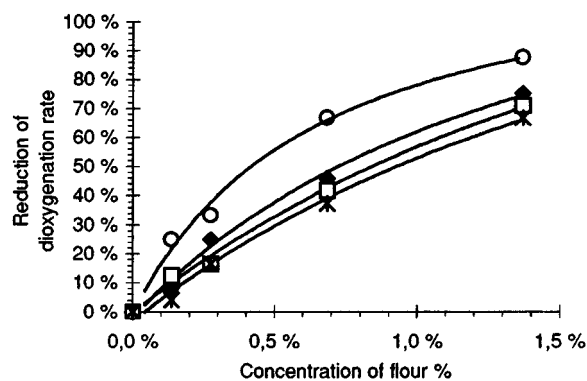
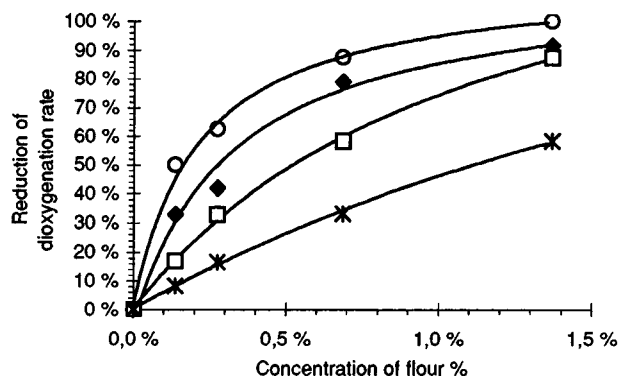
at the optimum pH (pH 6.0 for barley and pH 7.5 for rye). However, we measured the dioxygenation activities at the constant pH of 6.0, in which the activity of rye was 50% of that of barley. Thus, the comparison of different cereals and their dioxygenation potentials is valid only in the defined milieu. In the oat flour only 5% of the dioxygenation activity of barley flour was present. When the grains were thermally denatured by autoclaving, no dioxygenation activity was found in any of the flours.

When extraneous lipoxygenase was added to the water suspension of native flours, an increase in the apparent dioxygenation activity was noticed as expected (Table 1). The activities after the addition of extraneous lipoxygenase were, however, well below the arithmetic sum of activities measured separately for the flours alone or corresponding amounts of soybean lipoxygenase in the absence of flour. The activity after supplementation was lowest in oat flour, which was evident as an 8.8 nkat extraneous lipoxygenase addition to the 0.41% oat flour–water suspension resulted in a total dioxygenation activity that was only 3.0 nkat. In other cereals the total activity after the same supplementation was 7.0–7.6 nkat. With 16.0 nkat of lipoxygenase, the total activities remained in the range 7.7–11.6 nkat. The measured activity was lowest in oat flour suspension even with the higher amount of extraneous lipoxygenase.

When extraneous lipoxygenase was added to autoclaved flours in the same ratios as above, the apparent dioxygenation activities were unexpectedly slightly higher than when lipoxygenase was added to native flours with endogenous lipoxygenase activities (Table 1). From the results shown in Table 1 it is evident that the total amount of lipoxygenase in the flour–water suspension does not determine the dioxygenation rate. Furthermore, a heat stable inhibitor of dioxygenation is present in flours.

Effect of Dosage of the Denatured Flours. The inhibition was further characterized by varying the concentration of denatured flours in water suspensions supplemented with the fixed amount of linoleic acid and lipoxygenase. As the amount of flour was increased, the inhibition of oxidation reaction was clearly enhanced. The effective range of flour concentrations was ~0.1–1.4% (w/v) (Figure 2). The oat flour turned out to be the most effective inhibitor and caused the oxidation rate to diminish by 50% when flour concentration was 0.41% (IC_{50%} concentration). Wheat, rye, and barley had a markedly larger IC_{50%} concentrations: 0.73%, 0.83%, and 0.93%, respectively.

Effect of Different Oat Fractions. Of the denatured flours, the oat flour was found to be the most effective inhibitor of dioxygenation. Therefore, it was

**Figure 2.** Effect of the dosage of denatured flours on the oxygenation of linoleic acid catalyzed by soybean lipoxygenase: oat flour (○), barley flour (◆), wheat flour (□), and rye flour (*).**Figure 3.** Effect of the dosage of different oat fractions on the oxygenation of linoleic acid catalyzed by soybean lipoxygenase: soluble fiber (○), fiber (◆), protein (□), and starch (*).

of interest to study whether an even more potent inhibitor could be found by fractionation of oat. Oat was fractionated into soluble fiber, fiber, starch, and protein fractions.

No intrinsic dioxygenation activity was detected in any of the processed oat fractions. However, the inhibitory properties prevailed but were unevenly distributed among different fractions. With equivalent amounts of different fractions the inhibition was in the order soluble fiber (IC_{50%} concentration = 0.16%) > fiber (0.27%) > protein (0.52%) > starch (1.1%) (Figure 3). The dioxygenation inhibitor was distinctly concentrated from the whole meal oat into the soluble fiber. This is evident as the IC_{50%} concentration for soluble fiber was less than half of that for unfractionated oat flour, while the IC_{50%} concentration for starch was twice that found in oat flour.

The inhibition appears to increase linearly as the flour concentration was increased. This linearity remained until the IC_{50%} concentration was reached (Figures 2 and 3). On the other hand, to depress the dioxygenation rate below the level of detection, >10 times the IC_{50%} concentration was needed. This was established only with the soluble oat fiber (Figure 3).

Fate of Supplemented Linoleic Acid in Flour–Water Suspensions. The different behavior of oat and its fiber fractions raised the question whether the oat flour–linoleic acid interaction was different from that of other cereals and whether such differences could explain the antioxidative properties. Therefore, the fate and distribution of extraneous linoleic acid, supplemented in aqueous buffer, were studied in different cereal slurries (Figure 1). The amount of linoleic acid

Table 2. Recovery of Supplemented Linoleic Acid (LA) from Cereal–Water Suspensions^a

| | LA remaining in the particle-free buffer (%) | LA bound ^b to flour particles (%) | recovery of LA (%) |
|-----------|--|--|--------------------|
| oat | 64 | 18 | 82 |
| oat fiber | 48 | 45 | 93 |
| barley | 17 | 6.8 | 24 |
| rye | 20 | 22 | 41 |
| wheat | 27 | 32 | 59 |

^a Flours (100 mg each) were mixed for 15 min with 7 mL of aqueous buffer containing 8.6 mg of linoleic acid, after which time the flour particles were separated from the buffer. ^b Obtained from the data shown in Table 3 by adding the amounts of LA in the buffer extract, in the solvent extract, and in the unextractable particle fraction.

remaining in the particle-free aqueous buffer after the buffer was mixed with flour sample was determined. The linoleic acid absorbed into the flour particles was further consecutively extracted first with neutral aqueous buffer and subsequently with organic solvent. The linoleic acid in these extracts as well as in the particles remaining from the solvent extraction was also determined. The sum of linoleic acid detected in the buffer extract, solvent extract, and remaining particle fraction represents the amount of linoleic acid absorbed in flour particles, while the total yield was obtained by adding together the linoleic acid amounts in each fraction. The contents of endogenous cereal linoleic acid were in every case subtracted from the corresponding assays.

Part of the supplemented linoleic acid remained in the buffer in which it was introduced to the flour sample while part of it was absorbed in flour particles. A notable portion of the linoleic acid did also vanish during the treatment, so that the total yield of supplemented linoleic acid varied between 24% and 93% with different flours (Table 2). From oat flour and oat fiber suspensions were obtained high recoveries, 82% and 93%, respectively. For barley, rye, and wheat flours the value varied between 24% and 59%, reflecting the presence of endogenous lipoxygenase (Table 1). The amount of vanished linoleic acid corresponded well with the measured lipoxygenase activity except for wheat and rye. Despite the fact that by measuring the oxygen consumption a slightly higher lipoxygenase activity was detected for the wheat than for the rye (Table 1), a higher amount of linoleic acid was recovered from the wheat flour suspension (Table 2). However, both of these values were well below the yield obtained in oat flour and oat fiber suspensions.

The amount of linoleic acid remaining in the particle-free buffer varied dramatically with different cereals, from 17% to 64% of the supplemented linoleic acid. The variation was resulted by the differences in both the lipoxygenase activities and the amounts of linoleic acid absorbed in flour particles. Interestingly the oat fiber, which was a more efficient inhibitor of the dioxygenation reaction than whole meal oat, did also absorb twice the amount of linoleic acid compared to whole meal oat. Moreover, the relatively high recovery of linoleic acid from the wheat flour suspension compared to its high lipoxygenase activity can also be linked to the observation that wheat flour absorbed 1.5 times the amount of linoleic acid compared to rye flour. It is evident that absorption of linoleic acid to insoluble material and its protection against oxygenation appear to correlate. Physical protection of linoleic acid may well represent the primary mechanism by which flours retard the dioxygenation. Results presented in Table 3 further

Table 3. Distribution of Extraneous Linoleic Acid (LA) Bound to Flour Particles into the Aqueous Buffer Extractable, the Solvent Extractable, or the Unextractable Fraction

| | LA extracted with buffer (%) | LA extracted with solvent (%) | unextractable LA (%) |
|-----------|------------------------------|-------------------------------|----------------------|
| oat | 3.5 | 13 | 1.8 |
| oat fiber | 7.6 | 37 | 0.9 |
| barley | 2.1 | 4.6 | 0.0 |
| rye | 2.7 | 19 | 0.0 |
| wheat | 3.0 | 29 | 0.8 |

Table 4. Amounts of Specified and Total Fatty Acids Present in the Suspensions of Barley Flour, Barley Flour plus Oat Flour, or Barley Flour plus Oat Fiber in the Beginning of the Heating Period

| | 16:0 (mg) | 18:1 (mg) | 18:2 (mg) | 18:3 (mg) | total fatty acids (mg) |
|--|-----------|-----------|-----------|-----------|------------------------|
| 5 g of barley flour | 31 | 18 | 73 | 6.5 | 133 |
| 5 g of barley flour + 1 g of oat flour | 40 | 34 | 86 | 6.5 | 172 |
| 5 g of barley flour + 1 g of oat fiber | 43 | 50 | 99 | 6.8 | 206 |

support such a conclusion. Though linoleic acid was introduced as aqueous buffer, only 10–33% of acid initially absorbed into the flour particles was recovered by washing the particles with the same buffer. The fact that organic solvent extractions were needed to liberate the acid suggests that relatively firm complexes between the linoleic acid and flour matrixes were spontaneously formed.

Effect of Oat Flour and Oat Fiber Supplementation on the Dioxygenation of Barley Flour–Water Suspension. As the oat flour and especially oat fiber were found to be most potent inhibitors of oxidation, their effect on the oxidation of barley flour–water suspension was studied. No extraneous linoleic acid or lipoxygenase was added. The barley flour–water suspension (10% w/v) was exposed to air at 80 °C in the presence of 0.3 mM FeSO₄, and the amounts of different fatty acids were measured during the incubation. The incubation was continued for 96 h, after which time <15% and <5% of the initial linoleic and linolenic acid, respectively, remained. As a comparison it may be noted that 74% of the palmitic acid could still be detected. During the incubation the ratio of linoleic to palmitic acid fell from 2.35 to 0.42. The specific loss of unsaturated acids indicates that significant oxidation had occurred, though the small decrease in the amount of palmitic acid suggests that also other processes, possibly the unspecific lipid deterioration or adsorption into vessel surface, take place.

Repetition of the assay in the presence of an additional 2% (w/v) oat flour or oat fiber increased the initial amounts of palmitic, oleic, and linoleic acid in suspension (Table 4). Oat flour supplementation had no significant effect on the progress of oxidation of the barley flour slurry. The ratio of linoleic to palmitic acid fell from 2.15 to 0.57, resembling the bare barley flour–water suspension. However, the addition of oat fiber retarded the oxidation, so that when the barley flour–water suspension was supplemented with 2% of the oat fiber, the ratio of linoleic to palmitic acid fell only from 2.30 to 1.46 (Figure 4).

DISCUSSION

The results of the present study have shown that an antioxidative effect in aqueous systems can arise from the addition of cereal flours. The protective effect

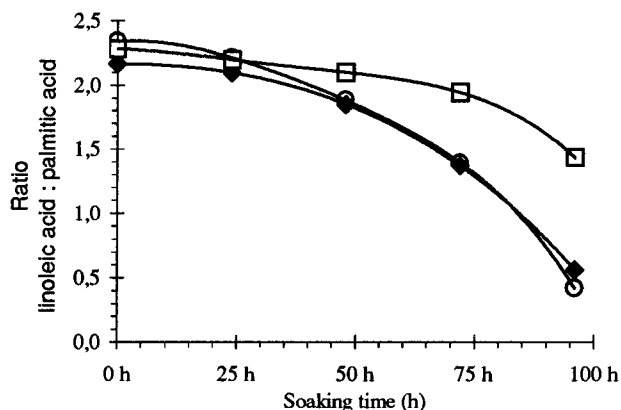


Figure 4. Effect of soaking time of barley flour (○), barley flour plus oat flour (◆), and barley flour plus oat fiber (□) suspensions on the ratio of linoleic acid to palmitic acid.

against oxidation of linoleic acid was established in a concentration as low as 0.1–1.4%. Oat was the most potent inhibitor of the studied cereals and caused an equivalent inhibition in 50% smaller concentration than other cereals. When oat was fractionated, the inhibitory properties were further concentrated into fiber and especially into soluble fiber fractions.

Under the experimental conditions used, the apparent dioxygenation activity was nearly linearly dependent of the amount of the lipoxygenase in the system provided no flour additions were used. However, when flours with endogenous lipoxygenase activities were added to the system, the dioxygenation rates were in fact reduced. Further, the total dioxygenation rates were unexpectedly increased when the endogenous lipoxygenase of the various flours was denatured.

The inhibition of dioxygenation described above is most likely to be the result of a range of diverse factors. Each flour suspension seems to have a characteristic apparent maximum rate that in practice cannot be increased by addition of moderate amounts of the catalyst. The reaction kinetics were typical for a system with substrate limitation. As linoleic acid was present in excess in flour slurries, the rate-determining step seems to be the availability of the substrate to the catalyst, rather than the total concentration of the substrate or the catalyst. However, a complete inhibition was not obtained with any of the flours tested, suggesting that an equilibrium exists under aqueous conditions between the available and the unavailable substrate. The described inhibition represents a retardation of dioxygenation under limited supply of substrate rather than actual intervention in the mechanism of dioxygenation itself. In any case, a dramatic protective effect on linoleic acid is obtainable especially by soluble oat fiber. The effective concentrations of flours were small enough, 0.1–1.4% to be considered as minor supplements.

Retardation of dioxygenation can be linked to the ability of cereals to absorb supplemented free linoleic acid into flour particles. The amount of absorbed linoleic acid varied with different cereals from 0.6% to 4.0% of flour (w/w), which is of the same order as the effective range causing the inhibition as determined by oxygen consumption measurements. Furthermore, the capacity of flour to absorb fatty acids was also of the same order that has been obtained for albumin. According to Duniec and Robak (1984), albumin was effective in reducing the oxidation rate when the amount of arachidonic acid was 0.8–3.3% of that of albumin.

Oat fiber, but not oat flour, was found to be able to retard oxidation of heated barley flour–water suspension. Oxidation was not completely inhibited by addition of oat fiber, but retardation was evident. The total amount of linoleic acid was $\approx 10\%$ of the oat fiber present in the suspension. It should be noted, however, that the amount of linoleic acid present as free acid or otherwise liable to oxidation is much smaller than the total amount of linoleic acid. On the other hand, Tian and White (1994) have shown that, at levels of 0.005%, methanol extracts of oat are capable of retarding the oxidation of cottonseed and soybean oil both as pure oil and as emulsion. However, the amount of oat needed to prepare the solvent extract is beyond any commercial application.

The phenomenon described in the present study is probably a major factor affecting the antioxidative capacity of cereal flours in aqueous systems and overrules the effect of phenolic compounds. Our laboratory is currently actively studying means by which the antioxidative effect can be stimulated in situations faced in food processing. In addition, concentration of the factor causing the reduction in the dioxygenation rate is in process.

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Received for review June 3, 1997. Accepted September 23, 1997.®

JF970458T

® Abstract published in *Advance ACS Abstracts*, November 1, 1997.