Food Chemistry **21** (1986) 145–152

Effect of Sequestering Agents on Lipid Oxidation in Cooked Meats

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(Received: 3 December, 1985)

ABSTRACT

To retard lipid oxidation, which affects the sensory and nutritional quality of cooked meats, a number of sequestering agents (ortho- and polyphosphates, citric acid, ethylenediaminetetraacetic acid (EDTA), diethylenetriaminepentaacetic acid (DTPA), salicylic acid, the sodium salts of oxalic, phytic and lactic acids, glycine, cysteine, 8-hydroxyquinoline and catechol) were added to ground pork prior to cooking. The TBA numbers of meat were evaluated after cooking (day 1) and after 7, 14, 21, 28 and 35 days of storage at 4°C by the 2-thiobarbituric acid (TBA) test. It was found that catechol, EDTA, DTPA, sodium pyrophosphate and, to a lesser extent, sodium tripolyphosphate, substantially lowered the TBA numbers while the other sequestering agents had only a marginal effect. Mixed-chelating systems composed of salicylic acid and either EDTA or DTPA did not show any synergistic effects.

Food Chemistry 0308-8146/86/$03·50

INTRODUCTION

A major cause of meat quality deterioration during storage is lipid oxidation. The rate of uptake of oxygen by a fatty compound increases with the degree of unsaturation, and, consequently, the meat is prone to develop a rancid flavour during storage (Wilson *et al.*, 1976). Whilst, in fat or oil, oxidation usually occurs through an autocatalytic process, in foods it becomes more complex. In meats, oxidative enzymes (Tappel, 1952; 1953), hematin compounds (Haurowitz *et al.*, 1941), traces of metals or metal ions, in particular, iron, copper and nickel (Ingold, 1962; Evans *et al.*, 1952), heat, light and salt (Watts, 1954; Chen *et al.*, 1984) generally influence the rate of lipid oxidation. Sequestrants or chelators combine with metal ions to form complexes which, depending on their stability, immobilize the metal ions in the system and therefore delay the onset of oxidation. Many of the chelators which may be used in food products occur naturally; these include polycarboxylic and hydroxycarboxylic acids, polyphosphates and amino acids.

In this paper we report on the use of a number of sequestrants to retard lipid oxidation in cooked pork.

MATERIALS AND METHODS

All chemicals used in these studies were reagent or food grade materials, and were used without any further purification. The levels used were, in general, 3000 ppm for phosphates and polyphosphates and 500 ppm for others, unless otherwise specified.

Fresh pork loin was deboned and trimmed to remove most of the surface fat. It was then ground twice using an Oster Meat Grinder, Model 990–68 (plate size 4·8 mm). Sequestrants were added directly to the meat followed by the addition of 20 % of distilled water. The mixture was then thoroughly homogenized.

The homogenized meat systems were cooked in a thermostated water bath for *ca.* 40 min to reach an internal temperature of 75 ± 1 °C, cooled to room temperature, and then each cooked meat system was re-homogenized in a Waring blender for 30 s and stored in a plastic bag at 4 °C until use. The oxidative state of the meat samples was determined after cooking (day 1) and after 7, 14, 21, 28 and 35 days of storage by a

modified version of the 2-thiobarbituric acid (TBA) procedure of Tarladgis *et al.* (1960).

In all cases, 10 g of sample was weighed on a nitrogen-free weighing paper and then transferred to a 500-ml round-bottomed flask containing 97·5 ml of distilled water, 2·5 ml of 4N HCl, a few drops of Dow Antifoam A and several glass beads. The mixture was then fractionally distilled and 50 ml of distillate was collected over a period of *ca.* 20 min.

In a vial containing 5 ml of distillate, 5 ml of 0·02M aqueous solution of 2-thiobarbituric acid was added. The vials were capped and heated in a boiling water bath for a period of 35 min, cooled to room temperature and the absorbance of the resultant pink-coloured chromogen was measured at 532 nm (maximum) using a Beckann DU-7 Spectrophotometer.

A solution of 1,1,3,3-tetramethoxypropane (TMP) was used as standard to obtain the conversion factor for absorbance values to TBA numbers.

RESULTS AND DISCUSSION

To convert the absorbance units to TBA numbers, defined as milligrams of malonaldehyde equivalent per kilogram of the mixture, a value of 8·1 was obtained using TMP standard solutions. This compares well with the value of 7·8 reported by Tarladgis *et al.* (1960).

Table 1 summarizes the TBA numbers of meat samples treated with non-phosphorus sequestrants and Table 2 gives the results for samples treated with phosphates and polyphosphates. The TBA values reported are expressed as the mean of triplicate samples $\pm$ standard deviation.

The TBA numbers generally increased with the length of storage after cooking. However, when the initial TBA numbers (day 1) were less than 0·5, they did not generally exceed 1·0 during the 5-week storage period. For untreated meat, the oxidation proceeded further, probably due to the formation of ferric hemoproteins (Younathan & Watts, 1959).

Citric acid and monoglyceride citrate had only a slight effect on retarding lipid oxidation, as indicated by the TBA numbers (Table 1). Olcott & Mattill (1936) reported that citric acid was ineffective in lard, although it acted as a powerful synergist with antioxidants, even when metals were not present in measurable quantities.

The antioxidant effect of 8-hydroxyquinoline was quite pronounced, giving a TBA value of 1·62 after 28 days of storage (Table 1). Catechol was even more effective.

TABLE 1
Effect of Sequestrants on the TBA Values of Cooked Pork During Storage at 4°C[a]

Sequestrant system	*Storage time (Days)*					
	1	*7*	*14*	*21*	*28*	*35*
No additive	4·95 ± 0·32	8·96 ± 0·73	10·5 ± 0·62	11·7 ± 0·86	12·1 ± 0·83	13·7 ± 0·92
Citric acid (500 ppm)	2·74 ± 0·24	8·08 ± 0·34	8·77 ± 0·42	10·0 ± 0·32	10·8 ± 0·51	12·6 ± 0·48
Monoglyceride citrate (500 ppm)	5·40 ± 0·32	8·57 ± 0·29	8·23 ± 0·43	9·92 ± 0·35	9·39 ± 0·72	9·16 ± 0·89
8-Hydroxyquinoline (500 ppm)	0·47 ± 0·05	0·74 ± 0·09	0·75 ± 0·06	0·94 ± 0·11	1·62 ± 0·08	—
Catechol (200 ppm)	0·09 ± 0·01	0·46 ± 0·05	0·46 ± 0·03	0·48 ± 0·02	0·66 ± 0·05	0·85 ± 0·03
Na_2EDTA (500 ppm)	0·31 ± 0·02	0·64 ± 0·03	0·71 ± 0·02	0·86 ± 0·05	0·90 ± 0·03	0·96 ± 0·08
DTPA (500 ppm)	0·29 ± 0·02	0·38 ± 0·02	0·33 ± 0·02	0·34 ± 0·01	0·33 ± 0·03	0·36 ± 0·02
Salicylic acid (1000 ppm)	4·12 ± 0·11	9·08 ± 0·42	9·52 ± 0·31	9·41 ± 0·42	9·68 ± 0·58	11·1 ± 0·62
Sodium oxalate (500 ppm)	4·74 ± 0·11	9·48 ± 0·32	11·5 ± 0·31	12·7 ± 0·58	13·7 ± 0·63	14·5 ± 0·68
Sodium phytate (500 ppm)	3·06 ± 0·32	7·24 ± 0·21	7·64 ± 0·22	9·09 ± 0·11	10·1 ± 0·38	9·74 ± 0·51
Sodium lactate (500 ppm)	4·05 ± 0·31	7·37 ± 0·51	9·00 ± 0·24	8·71 ± 0·33	9·01 ± 0·30	11·6 ± 0·38
Glycine (500 ppm)	4·69 ± 0·14	7·32 ± 0·18	8·61 ± 0·16	9·66 ± 0·32	12·0 ± 0·23	13·3 ± 0·31
Cysteine (500 ppm)	2·63 ± 0·30	7·16 ± 0·28	6·60 ± 0·38	9·04 ± 0·50	12·0 ± 0·18	11·5 ± 0·61
Salicylic acid (1000) + Na_2EDTA (500 ppm)	0·58 ± 0·06	1·02 ± 0·11	1·59 ± 0·09	1·26 ± 0·13	1·31 ± 0·06	—
Salicylic acid (1000) + DTPA (500 ppm)	0·44 ± 0·05	0·42 ± 0·07	0·35 ± 0·03	0·35 ± 0·03	0·38 ± 0·02	—

[a]The cooked meats contained 72·4 ± 0·2% water and 10·5 ± 0·2% fat (AOAC, 1980).

TABLE 2
Effect of Phosphates and Polyphosphates on the TBA Values of Cooked Pork During Storage at 4°C

Sequestrant system	*Storage time (Days)*					
	1	*7*	*14*	*21*	*28*	*35*
No additive	4·95 ± 0·32	8·96 ± 0·73	10·5 ± 0·62	11·7 ± 0·86	12·1 ± 0·83	13·7 ± 0·92
Monosodium phosphate (3000 ppm)	5·60 ± 0·23	10·6 ± 0·48	10·5 ± 0·30	10·1 ± 0·81	10·7 ± 0·53	11·0 ± 0·38
Disodium phosphate (3000 ppm)	4·56 ± 0·21	9·19 ± 0·13	9·58 ± 0·43	9·89 ± 0·62	10·5 ± 0·49	9·20 ± 0·30
Sodium hexametaphosphate (3000 ppm)	1·41 ± 0·05	3·46 ± 0·11	4·70 ± 0·09	6·00 ± 0·23	7·88 ± 0·18	8·78 ± 0·31
Sodium tripolyphosphate (1500 ppm)	1·57 ± 0·11	3·33 ± 0·22	3·63 ± 0·12	4·39 ± 0·09	4·28 ± 0·05	4·95 ± 0·25
Sodium tripolyphosphate (3000 ppm)	0·31 ± 0·01	0·58 ± 0·02	0·90 ± 0·04	1·05 ± 0·05	1·39 ± 0·09	2·07 ± 0·04
Sodium pyrophosphate (1500 ppm)	0·26 ± 0·01	0·30 ± 0·01	0·51 ± 0·02	1·64 ± 0·03	2·14 ± 0·08	3·41 ± 0·06
Sodium pyrophosphate (3000 ppm)	0·35 ± 0·01	0·32 ± 0·01	0·39 ± 0·02	0·48 ± 0·01	0·69 ± 0·01	1·13 ± 0·03
Sodium tripolyphosphate (1500 ppm) + Sodium pyrophosphate (1500 ppm)	0·16 ± 0·02	0·14 ± 0·01	0·18 ± 0·02	0·33 ± 0·01	0·65 ± 0·03	1·10 ± 0·05

[a] The cooked meats contained 72·4 ± 2% water and 10·5 ± 2% fat (AOAC, 1980).

Of the other chelating agents, the disodium salt of ethylenediaminetetraacetic acid (Na_2EDTA) was found to be highly effective and the TBA numbers did not exceed 1·0 over the 5-week storage period. EDTA is a well established metal sequestering agent (Hole, 1980) and chelates divalent metals, particularly free iron that is endogenous to the muscle or released on cooking. EDTA also has an advantage in that it causes a substantial delay in the outgrowth of *C. botulinum* in nitrite-treated meats (Tompkin *et al.*, 1978). Diethylenetriaminepentaacetic acid (DTPA), with more binding sites for chelation than EDTA, was even more effective in retarding lipid oxidation and TBA values reached only 0·36 after 5 weeks of storage (Table 1).

Salicylic acid (SA) alone had little or no effect on TBA values (Table 1). It has also been shown to have no effect on the removal of plutonium or cadmium from the tissue of mice and, while the effect of EDTA and DTPA was minor (Schubert & Derr, 1978), mixed ligands of EDTA–SA and DTPA–SA successfully removed both toxic chemicals. In fact, the chelating ability of these mixtures exceeded that of EDTA alone by at least 10^{13}. Based on these data we determined the TBA numbers of meats treated with these mixed ligands. However, results (Table 1) do not indicate any synergistic effect due to the use of these substances in combination.

The sodium salts of oxalic, phytic and lactic acids (Table 1) were only marginally effective for retarding lipid oxidation. The amino acids, cysteine and glycine, also had only a marginal antioxidant effect. This is in accord with the results of Sato & Hegarty (1971) for meats treated with cysteine, although this reagent is known to markedly accelerate rancidity when dissolved in water in contact with fats low in tocopherols (Watts, 1950).

All phosphate and polyphosphate chelators showed some antioxidant effects in the cooked meat (Table 2). Sodium tripolyphosphate (STPP) was reasonably effective, while sodium pyrophosphate (SPP) was better. They also showed some synergism in combination. Sodium hexametaphosphate (SHMP) and monosodium and disodium phosphate were much less effective. Tims & Watts (1958) have shown that only pyro-, tripoly- and hexametaphosphates protect cooked meats from oxidation; however, we did not find hexametaphosphate effective. In addition to antioxidant properties, phosphates, particularly sodium tripolyphosphate, have been shown to improve the colour, flavour and tenderness of meats (Smith *et al.*, 1984). Savich & Jansen (1954) reported that the colour

of ground meat was stabilized for extended periods when phosphates were added. Their influence on flavour has been attributed to the retention of proteins (Ellanger, 1972) and reduction of oxidative rancidity (Ramsey & Watts, 1963). The antioxidant rôle of polyphosphates is due to their ability to sequester heavy metals (Watts, 1950; Tims & Watts, 1958), particularly ferric ion, which are the major pro-oxidants in meat systems (Love & Pearson, 1974). Polyphosphates also improve the cooking yield of meats by increasing their water-holding capacity, due to the unfolding of the proteins in meat, by raising the pH and by a more specific phosphate effect (Mahon *et al.*, 1970).

In conclusion, it was found that catechol, EDTA, DTPA, sodium pyrophosphate and, to a lesser extent, sodium tripolyphosphate, substantially lower the fat oxidation, as indicated by the TBA numbers. The effect of other sequestrants was only marginal. Amongst these, polyphosphates, and perhaps EDTA, are obviously the chemicals of choice.

ACKNOWLEDGEMENTS

This work was supported by Agriculture Canada through Contract No. 0151-1-1429. Contribution No. 659 from the Food Research Institute.

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