

EFFECTS OF SOME ADDITIVES ON MUTAGEN FORMATION DURING FRYING OF GROUND BEEF

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Summary

2-Amino-3-methyl-imidazo[4,5-f]quinoline (IQ), 2-amino-3,4-dimethylimidazo[4,5-f]quinoline (MeIQ), 2-amino-3,8-dimethyl-imidazo[4,5-f]-quinoxaline (MeIQx) and 2-amino-3,4,8-dimethylimidazole[4,5-f] quinoxaline (4,8-DiMeIQx) are potent mutagens found in beef extract and fried beef. The effects that different food additives have on the formation of these mutagens during frying of ground beef were investigated. The food additives utilized included bisulfite, nitrite, phosphate, citrate, ascorbic acid, vitamin E and liquid smoke. Results demonstrated that all of the food additives studied inhibited formation of meat mutagens in variable amounts at different concentrations, except for pyrophosphate at 10ppm which was ineffective. Bisulfite at 10ppm was the most effective reducer of mutagens and was followed in order by liquid smoke (100ppm), ascorbic acid (100ppm), vitamin E (10% of fat), nitrite (10ppm) and citric acid (100ppm).

INTRODUCTION

MeIQ and MeIQx were originally isolated from broiled fish (Kasai et al., 1980a,b). MeIQx (Kasai et al., 1981) and 4,8-DiMeIQx (Knize et al., 1985) were isolated from beef. These compounds, which are classified as IQ-like compounds, were also present in beef extract and fried beef (Chen et al., 1990). We showed earlier that several of the synthetic antioxidants (BHA, PG and TBHQ) decreased the formation of these IQ-like compounds in fried ground beef (Pearson et al., 1992a). It has been proposed that free radical reactions may be involved in the formation of these meat mutagens (Pearson et al., 1992b). The synthetic antioxidants can scavenge free radicals and block the formation of the IQ-like compounds. However, not all antioxidant activity is due to the effects of free-radical termination. Thus, we have added different food additives to ground beef and determined their effects on the IQ-like compounds formation during frying.

MATERIALS AND METHODS

Methods
Ground beef was prepared to contain one of the following food additives: bisulfite (10 and 100ppm), nitrite (10 and 100ppm), pyrophosphate (10 and 100ppm), citrate (10 and 100 ppm), ascorbic acid (10 and 100 ppm), vitamin E (1 and 10% of the fat content) and liquid smoke

(100 and 1000 ppm). The ground beef was made into 100g, 0.5cm thick patties, which were fried in a frying pan set at 215 C for 9 min. per side. The fried patties were extracted using modifications of the methods of Bjeldanes et al. (1982), Hayatsu et al. (1983) and Felton et al (1984). A Versapack C18 column with 1M ammonium sulfate in methanol:ethanol:water (35:6:59, v/v/v) was used as the mobil phase in the HPLC analysis.

RESULTS AND DISCUSSION

Table 1 shows the effects of adding different food additives upon the formation of the IQ-like compounds. MeIQ was not detected in any of the samples. IQ, MeIQx and 4,8-DiMeIQx were detected in the control patties at 1558, 5028 and 730 ng/g of meat equivalents, respectively.

Sulfiting agents and ascorbic acid are reducing agents and function by transferring hydrogen atoms, thus serving as oxygen scavengers. Sodium bisulfite significantly inhibited formation of IQ and MeIQx at both concentrations ($P < 0.0005$). Bisulfite also inhibited the formation of 4,8-DiMeIQx at 10 ppm ($P < 0.025$) but had no statistically significant effect at 100 ppm ($P < 0.2$). Ascorbic acid inhibited formation of IQ, MeIQx and 4,8-DiMeIQx, but was not as effective as bisulfite. At 100 ppm, ascorbic acid inhibited IQ and MeIQx ($P < 0.025$ and $P < 0.01$, respectively) but had no statistically

Table 1. Effects of Different Food Additives on the Formation of IQ-Like Compounds During Frying of Ground Beef.

TREATMENT		IQ Mean values	MeIQx (ng/g of meat)	4,8-DiMeIQx equivalents)
CONTROL		1558	5028	730
Bisulfite	10 PPM	110	1272	17
Bisulfite	100 PPM	212	2127	494
Nitrite	10 PPM	1534	2077	204
Nitrite	100 PPM	214	2734	141
Pyrophosphate	10 PPM	436	2373	4958
Pyrophosphate	100 PPM	464	3627	370
Citric Acid	10 PPM	347	3760	121
Citric Acid	100 PPM	152	1695	709
Ascorbic Acid	10 PPM	178	1452	657
Ascorbic Acid	100 PPM	345	2464	290
Vitamin E	1% Fat	206	2299	493
Vitamin E	10% Fat	282	2094	0
Liquid Smoke	10 PPM	177	2033	35
Liquid Smoke	100 PPM	183	2232	202

significant effect on formation of 4,8-DiMeIQx ($P < 0.25$). At 1000 ppm, ascorbic acid significantly inhibited formation of IQ and MeIQx ($P < 0.005$ and $P < 0.05$, respectively), but caused only a slight inhibition of 4,8-DiMeIQx ($P < 0.1$).

Both sodium bisulfite and ascorbic acid were more effective in blocking mutagen formation at low levels than at high levels. On adding sodium bisulfite, the total meat mutagens detected amounted to 1399 ng/g (meat equivalent) at 10 ppm but 2833 ng/g (meat equivalent) at 100 ppm. For ascorbic acid, the total amount of meat mutagens amounted to 2287 ng/g (meat equivalent) at 100 ppm and 3099 ng/g at 1000 ppm. The reason that the lower levels were more effective than higher levels is not clear. One possible explanation is that different pHs may effect the amount of mutagens formed. Nitrite and liquid smoke also showed a similar tendency in the present study, with higher concentrations producing more nitrite-like compounds than lower concentrations. However, the effects of high concentrations of sodium bisulfite and ascorbic acid on mutagenicity were more pronounced than those of nitrite and liquid smoke.

Nitrite significantly inhibited IQ, MeIQx and 4,8-DiMeIQx at both 10 and 100 ppm. Both concentrations of nitrite were about equally effective in blocking mutagen formation. In the present study, at 100 ppm of added liquid smoke, formation of IQ ($P < 0.005$), MeIQx ($P < 0.005$) and 4,8-DiMeIQx ($P < 0.025$) was significantly inhibited. At 1000 ppm, liquid smoke inhibited formation of IQ ($P < 0.0005$), MeIQx ($P < 0.025$) and 4,8-DiMeIQx ($P < 0.05$).

Both citric acid and polyphosphates are chelating agents, which complex with prooxidant metal ions such as iron and copper thus, chelating agents can block the catalysis of lipid oxidation (Dziezak, 1986). At 10 ppm, sodium citrate significantly inhibited formation of IQ and 4,8-DiMeIQx ($P < 0.005$ and $P < 0.025$, respectively) but had little effect upon formation of MeIQx ($P < 0.2$). At 100 ppm, sodium citrate significantly inhibited both IQ and MeIQx ($P < 0.0005$ and $P < 0.005$, respectively), but there was no great difference ($P > 0.25$) in the concentration of 4,8-DiMeIQx in the control and the citrate treated samples.

It has been reported that short chain polyphosphates, for example pyrophosphate, are better heavy metal sequestrants than long chain polyphosphates (Steinhauer, 1983). In the present study, 100 ppm sodium pyrophosphate slightly inhibited formation of IQ, MeIQx and 4,8-DiMeIQx ($P < 0.1$). At 10 ppm, however, sodium pyrophosphate caused a small increase in formation of 4,8-DiMeIQx ($P < 0.1$), but it had no great effect on formation of IQ and MeIQx in comparison to the control.

Reducing agents (bisulfite and ascorbic acid) are more effective in inhibiting meat mutagen formation than chelating agents (sodium citrate and sodium pyrophosphate).

Tocopherol mixture (Cavitol) was also tested in the present study using two

concentrations (1 and 10% of fat content). The results indicate that 1% of mixed tocopherols significantly reduced IQ formation ($P < 0.0005$), but at 10% was less effective ($P < 0.1$). The tocopherols also inhibited formation of MeIQx at both concentrations tested ($P < 0.05$ for 1 % and $P < 0.01$ for 10%). Addition of tocopherols at a concentration of 1% slightly inhibited formation of 4,8-DiMeIQx ($P < 0.2$), but at 10% completely prevented its formation.

Results of the present study suggest that the compounds tested may stabilize the sugar fragment or react with the free radicals formed in the earlier stages of the browning reaction, and thus indirectly inhibit the formation of the meat mutagen precursors.

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