Effect of preheating and different solvents on antioxidant and antimicrobial activity of garlic in pork patties

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Abstract

Garlic of the Allium family contains a number of sulfur and phenolic compounds, which might have excellent antioxidant and antimicrobial activity in processed meats. This study was performed to evaluate antioxidant and antimicrobial activity of garlic, as affected by preheating and different extraction solvents, and its application to ground meat if it may improve functional properties. Natural components from garlic were extracted with water or methanol, and preheating effect on the antioxidative activity was also observed. Total phenolic content, DPPH-radical scavenging ability, iron chelating ability, reducing power and antimicrobial activity against *E. coli* O157:H7 and *Listeria monocytogenes* for garlic extracts were evaluated. Preheated garlic with methanol extraction (HGM) and fresh garlic with water extraction (FGW) showed the higher antioxidant and antimicrobial activity than the counterparts. The effect of HGM and FGW alone or in combination on antioxidative and antimicrobial activity in pork patties were evaluated. HGM alone and in combination with FGW inhibited the lipid oxidation of pork patties during storage, and the combination suppressed the microbial growth of *Enterobacteriaceae* in pork patties. Thus, the combination of FGW and HGW had better antioxidant and antimicrobial activity than single addition.

Introduction

Garlic is one of the most commonly used ingredients for several foods as a flavor enhancer, and contains a lot of sulfur and phenolic compounds (Lanzotti, 2006), which have excellent antioxidant and antimicrobial activity (Sallam et al., 2004). However, processing of garlic, especially heat treatment, might change its components, and consequently either increase or decrease the functionality of garlic. To date, not many studies have been performed to investigate the effect of preheating and different solvents on lipid oxidation and microbial growth of garlic in ground meat. Therefore, the objectives of this study were to evaluate the effects of preheating and different solvents on antioxidant and antimicrobial activity of fresh garlic and determine its application to ground meat products.

Materials and methods

Experiment 1. Fresh garlic was purchased from local market, peeled and washed. One batch of garlic was heated at boiling water (100^{o}C) for 30 min. After cooling at room temperature, the batch was freeze-dried together with the rest batch. Natural components from dehydrated garlic were extracted with water or methanol (1:10) at room temperature for 24 hr using magnetic stirrer. Antioxidant activity of each garlic extract was determined using total phenolic content, DPPH-radical scavenging activity, iron chelating ability and reducing power. Antimicrobial activity was determined by measuring the area of inhibition zone against *E. coli* O157:H7 and *Listeria monocytogenes*. Briefly, metal borer was used to make wells. *E. coli* O157:H7 or *Listeria monocytogenes* of about 10^{7} cfu/mL of TSB containing 0.75% agar was poured upper solidified TSB agar (1.5%) into petri dish. Wells were filled with 180 uL of garlic extracts. The plates were incubated for 48 h at 37^{o}C. Data were analyzed via one-way analysis of variance (ANOVA) with SPSS 14.0. Means were separated using Duncan's multiple range test.

Experiment 2. Fresh pork hams were purchased from a wholesale meat market, and trimmed to remove excess fats and connective tissues. Pork patties were manufactured with the following formulation: control patty (without ingredient); reference patty (with BHT 0.01%); patty 1 (with fresh garlic water extracts (FGW), 0.5%); patty 2 (with heated garlic methanol extracts (HGM), 0.5%); patty 3 (with FGW 0.5% + HGM 0.5%). The proportions of pork lean, fat and salt were 78.5, 20.0 and 1.5%, respectively. Pork patties were aerobically packaged using polystyrene plate. pH, hunter color, TBARS (Sinnhuber and Yu, 1977), total bacterial counts and *Enterobacteriaceae* of pork patties were measured at 0, 3, 7, 10, 14, 21 and 28 day of refrigerated storage. Data were analyzed via two-way ANOVA with SPSS 14.0, as factors for treatments (various ingredients) and storage. Means were separated using Duncan's multiple range test.

Results and discussions

Experiment 1. Effect of solvents and heating on the antioxidative and antimicrobial activit: Water

extraction showed higher yield than methanol counterpart ($P < 0.05$), whereas, no differences were found by heat treatment (Table 1). Pre-heating increased total phenolic content in methanol extracted garlic, whereas, reverse trend was observed in water extracted one ($P < 0.05$). HGM showed higher DPPH-radical scavenging activity and reducing power, but lower iron chelating ability than FGW (Fig. 1). FGW formed inhibition zone against *E. coli* O157:H7 (Fig. 2). These results were similar to those of Gazzani et al. (1998) who reported that antioxidant activity of garlic increased after heat treatment. Banerjee and Sarkar (2003) reported that aqueous extracts of garlic possessed a potent antimicrobial activity, as well. Therefore, HGM was selected as a better radical scavenger and reducing agent, and FGW as a better metal chelater and antimicrobial agent.

Table 1. Yield and total phenolic content of garlic extracts as affected by heating and solvent

	Fresh garlic		Heated garlic	
	Water	Methanol	Water	Methanol
Yield (%, dry base)	70.72[a]	5.46[b]	69.53[a]	4.83[b]
Total phenolic content (g GAE/100g dry weight of extracts)	1.21[b]	1.50[b]	0.51[c]	2.65[a]

[a-c] Means with same superscripts within same row are not different ($P > 0.05$).

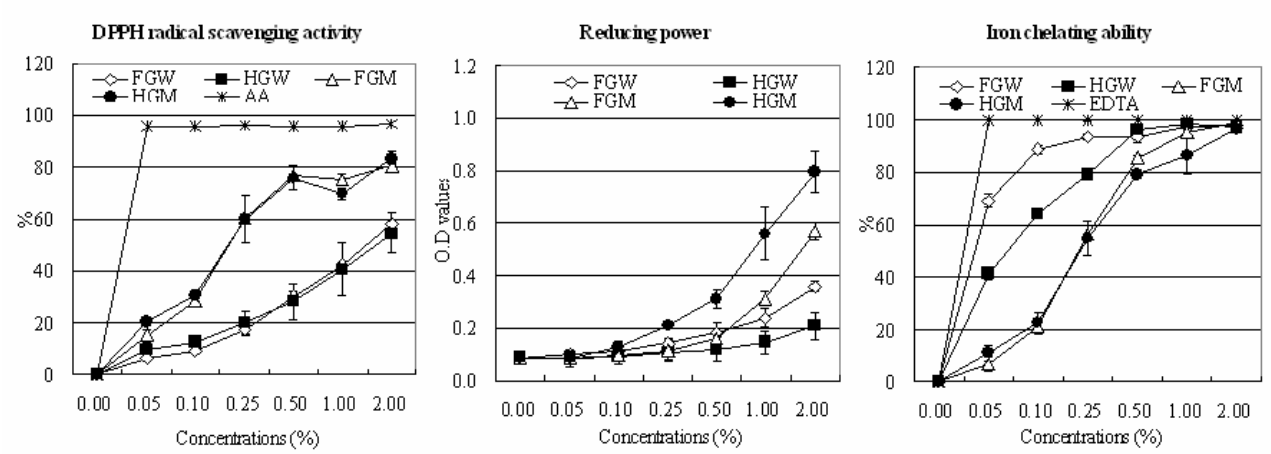

Figure 1. Effect of pre-heating, solvent and concentration of garlic extracts on antioxidant activities. FGW = fresh garlic with water extraction; HGW = heated garlic with water extraction; FGM = fresh garlic with methanol extraction; HGM = heated garlic with methanol extraction; AA = ascorbic acid; EDTA = ethylenediaminetetraacetic acid.

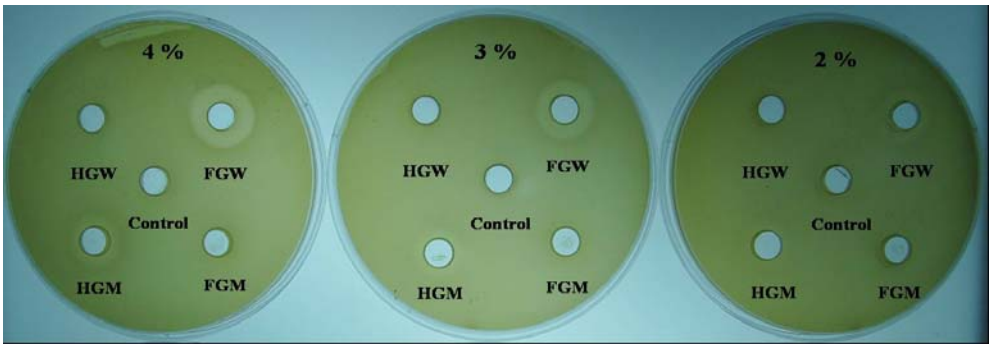

Figure 2. Effect of pre-heating, solvent and concentration of garlic extracts on antimicrobial activity. FGW, HGW, FGM, HGM are same as Figure 1. Control = sterilized water.

Experiment 2. Effects of HGM and FGM alone or in combination on antioxidative and antimicrobial activity: The effects of HGM and FGW alone or in combination on antioxidative and antimicrobial activity in pork patties were evaluated and are shown in Table 2. Most parameters were affected by treatment and storage time. pH and a (redness) values were decreased with the addition of garlic extracts, whereas, b (yellowness) values were increased with the addition of garlic extracts, regardless of type. HGM alone or in combination with FGW decreased TBARS and especially, the combination had an antioxidant activity as effective as butylated hydroxytoluene (BHT). FGW alone or in combination with HGM suppressed the microbial growth of *Enterobacteriaceae* in pork patties. Most parameters increased with increased storage time, except for hunter a value. Thus, the combination of FGW and HGW had better antioxidant and antimicrobial activity than single addition.

Table 2. Effect of treatment and storage time on pH, color (L, a, b), thiobarbituric acid reactive substance (TBARS) and microbial (TPC, VRB) counts of pork patties during storage

Main effect	pH	L	a	b	TBARS	TPC	VRB
Treatment	*	NS	*	*	*	**	**
Storage	*	*	*	*	*	*	*
Treatment * Storage	*	NS	*	NS	*	NS	NS
Treatment							
CTL1	5.96^{A}	60.47	6.69^{B}	7.56^{C}	2.12^{A}	4.86^{A}	4.43^{A}
CTL2	6.01^{A}	60.29	7.10^{A}	7.48^{C}	0.77^{C}	4.75^{A}	4.41^{A}
TRT1	5.90^{B}	61.12	4.97^{C}	8.11^{B}	1.97^{A}	4.51^{AB}	3.94^{B}
TRT2	5.88^{B}	61.22	3.51^{D}	9.34^{A}	1.55^{B}	4.62^{A}	4.06^{AB}
TRT3	5.84^{B}	61.43	3.21^{D}	9.63^{A}	0.97^{C}	4.06^{B}	3.55^{B}
Storage (days)							
0	5.73^{c}	58.04^{d}	8.79^{a}	7.93^{cd}	0.22^{e}	2.84^{c}	2.00^{e}
3	5.76^{c}	58.74^{d}	6.46^{b}	7.40^{de}	0.46^{e}	2.80^{c}	2.11^{e}
7	5.78^{c}	60.42^{c}	4.99^{c}	7.21^{e}	0.93^{d}	3.21^{c}	2.64^{de}
10	5.77^{c}	60.78^{bc}	4.35^{d}	7.85^{cd}	1.14^{cd}	3.85^{c}	3.49^{d}
14	5.79^{c}	61.83^{b}	3.69^{de}	8.42^{c}	1.42^{c}	5.50^{b}	4.96^{c}
21	6.10^{b}	63.14^{a}	3.36^{ef}	9.49^{b}	2.70^{b}	6.48^{ab}	6.21^{b}
28	6.50^{a}	63.39^{a}	4.03^{f}	10.65^{a}	3.47^{a}	7.23^{a}	7.16^{a}

$^{A-D}$Means with same letters within same column (treatment) are not different ($P > 0.05$). $^{a-f}$Means with same letters within same column (storage time) are not different ($P > 0.05$). CTL1 = control patty with salt 1.5%; CTL2 = reference patty with BHT 0.01% + salt 1.5%; TRT1 = patty with fresh garlic water extracts (FGW) 0.5% + salt 1.5%; TRT2 = patty with heated garlic methanol extracts (HGM) 0.5% + salt 1.5%; TRT3 = patty with FGW 0.5% + HGM 0.5% + salt 1.5%; TBARS = thiobarbituric acid reactive substance; TPC = total bacterial counts; VRB = *Enterobacteriaceae*; NS = not significant; * = $P < 0.001$; ** = $P < 0.05$.

Conclusions

Antioxidative and antimicrobial activity of garlic was affected by heat treatment and different extraction solvents. HGM was higher radical scavenger and reducing agent, and lower metal chelater and antimicrobial agent than FGW. The combination of FGW and HGM had better antioxidative and antimicrobial activity than single addition in pork patties.

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