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Preliminary study of superworm (*Zophobas morio*) larvae oil for antioxidant and antimicrobial activities in ground pork (#182)

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Introduction

Synthetic preservatives, which currently being used to reduce microbial growth and thereby extend the shelf-life of meat products [1], but affects the health of consumers. The many reports has been focused on extracts from medical plants and used to improve the sensory characteristics and shelf-life of foods [2]. In the other hand, those meat products have an unpleasant odor, which according to the smell of those plants. However, the products still have smell of original material. Insect oil are total polyunsaturated fatty acids in insects content may be up to 70% of total fatty acids [3] may be effect to the stability of oxidation reaction. Therefore, the objective of this study was to potential of insect oil from *Z. morio* to antioxidant and antimicrobial activities, and the application of insect oil in ground pork as a natural preservative.

Methods

Raw materials and preparation: The larvae of superworm were reared by using KMITL-P21-21 formulation as food. The 70-days old of larvae were cleaned and oven-dried at 60°C, and grounded into fine pieces. The dried insect was subjected to 95% ethanol (1:3) by maceration method and heating on a stirring hot plate at 65°C for 1 h. The solution was then filtered under a vacuum through filter paper. After filtering, the ethanol solvent was removed by rotary evaporation, to obtain the insect oil.

Determination of in vitro: DPPH and ABTS radical scavenging activity was determined using the method described by [4] and [5], respectively. Antimicrobial activity against pathogenic bacteria was performed using agar well diffusion method [6].

Application of insect oil in ground pork: pH levels were determined according to [7]. The colour *values* (L*, a*, and b*) of the raw ground pork meat fillets was determined using a Colorimeter Mini Scan EZ 4000L (USA). Thiobarbituric acid reactive substances (TBARS) value was determined according to [8]. Observation was during storage (4°C) for 0, 30, 60 and 90 days after treatment.

Results

In vitro: The results revealed that antioxidant activities of oil insect were lower than BHT and a-tocopherol. The IC_{50} from insect oil using DPPH and ABTS assays were approximately 0.14% and 0.16%, respectively. The lower IC_{50} values of BHT and a-tocopherol measured by DPPH assay (0.0002 and 0.0036%, respectively) and ABTS assay (0.0038 and 0.0017, repectively) were observed (Table 1). The insect oil showed non-inhibition all target microorganisms as diameter of inhibition zone <10.0 mm (Table 2).

In vivo: It was found that oil insect had no effect on pH and colour of ground pork compared to control and BHT groups (Table 3). The TBARS method has been used to determine the degree of lipid oxidation through MDA compound. The results showed that the lowest MDA content was observed in minced pork containing BHT. However, total MDA content in minced pork adding oil insect was lower than control ($p<0.05$) (Table 3).

In addition, Ground pork containing oil insect (52.65%) was higher ($p>0.05$) DPPH scavenging activity than control (41.25%) and 0.1%BHT (40.69). In the same way, ground pork containing oil insect showed higher ($p<0.05$) ATBS radical cation decolorization than control and 0.1%BHT as shown in Table 3.

The effective of 10% insect oil to antioxidant activity in this study were lower than the standard groups. While, the insect oil gave higher than fruit wines [9] and spice extracts [6] were previous studies. The IC_{50} of these methods exhibited high activity in accordance with total phenolic content in fruit plants were high [9]. Whereas, total polyunsaturated fatty acids in insects content may be up to 70% of total fatty acids [3] may be effect to the stability of oxidation reaction. The result found that the insect oil from superworm could not use to antibacterial in meat products. These results suggest that these antioxidants from insect oil could not delayed lipid oxidation during storage, but have a tendency antioxidant more than negative group. Although antioxidant activity is reported to be associated with phenolic compounds, a possible synergism between phenolic compounds and other compounds might be responsible for these observations, especially total polyunsaturated fatty acids in insect oils.

Conclusion

The IC_{50} from oil insect using DPPH and ABTS assays were higher value than positive control (BHT and a-tocopherol), which indicated lower antioxidant activity. Antibacterial activity of oil insect against pathogenic bacteria was not observed. This insect oil was applied to ground pork. It was found that meat sample containing oil insect revealed higher antioxidant activity. In addition, total MDA content in minced pork after adding oil insect was lower than control during storage time until 90 days. This finding could be explained that adding oil insect in minced pork enhanced the antioxidant activity and could be against lipid peroxidation during storage.

Notes

References

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Samples	Anioxidant Methods	
	DPPH (IC_{50})	ABTS (IC_{50})
superworm larvae (*Z. morio*)	0.14[a1/]	0.16[a]
BHT	0.0036[b]	0.0038[b]
α-tocopherol	0.0002[b]	0.0017[b]

[1/] Means in column followed by the same letter are not significantly different (P < 0.05)

Parameters	Determination of *in vivo* antioxidant activity		
	control	0.01% BHT	0.5% insect oil
pH	6.49[a1/]	6.50[a]	6.48[a]
colour values			
L*	54.74[a]	53.74[a]	53.54[a]
a*	4.41[a]	4.05[a]	4.34[a]
b*	12.89[a]	12.40[a]	13.24[a]
TBARS values (mg/kg meat)			
day 0	0.33[a]	0.34[a]	0.35[a]
day 30	0.56[a]	0.38[a]	0.50[a]
day 60	0.66[a]	0.41[c]	0.61[b]
day 90	1.08[a]	0.43[c]	0.91[b]
DPPH and ABTS radical scavenging activity			
DPPH (%activity)	41.25[b]	40.69[b]	52.65[a]
ABTS (%activity)	68.45[a]	83.73[a]	88.14[a]

[1/] Means in column followed by the same letter are not significantly different (P < 0.05)

Table 3.
The effects of 0.5% insect oil from superworm larvae on pH, colour values, TBARS values, and DDPH and ABTS radical decolorization in ground pork.

Table 1.
The percentages of DDPH and ABTS radical decolorization in insect oil from superworm larvae.

Pathogenic Microoraganisms		Diameter of Inhibition Zone (mm)
control		< 10.0
bacterial standards	*Staphylococcus aureus* TISTR 118	< 10.0
	Pseudomonas aeruginosa ATCC 9027	< 10.0
	Salmonella enterica serovar	< 10.0
	Enteritidis DMST 17368	< 10.0
	Escherichia coli ATCC 8739	< 10.0
	Pseudomonas fluorescens JCM 5963[T]	< 10.0
in chicken meats	*Escherichia coli*	< 10.0
	Staphylococcus aureus	< 10.0
	Salmonella spp.	< 10.0
	Citrobacter freundii	< 10.0
in pork meats	*Salmonella*	< 10.0

Table 2.
Antibacterial activities of 10% insect oil from superworm larvae for chosen pathogenic microoraganisms.

Notes