

Aktivitas Antelmintik Infusa Daun Kemangi (*Ocimum sanctum* Linn.) Terhadap *Ascaris suum* Secara In Vitro

Anthelmintic Activity of Basil Leaves (*Ocimum sanctum* Linn.) Infusion Against *Ascaris suum* In Vitro

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Abstract

Ascaris suum is a parasite nematode that causes infection in swines with high prevalence rates in host populations and usually associated with liver damages called “milk spots” caused by larvae migration, resulting in organ condemnation. Basil leaves (*Ocimum sanctum*) phytochemical constituents contains flavonoid, phenol and tannin. Tannins and phenolics are known to interfere with the energy generation in helminth parasites by uncoupling oxidative phosphorylation and also bind to free proteins in the gastrointestinal tract of host animal or glycoprotein on the cuticle of the parasite and leading to death. This study was aimed to determine the activity of basil leaves (*Ocimum sanctum*) infusion in several concentrations against *A. suum* *in vitro*. This research used six treatments and four replications. This research used 10 *A. suum* in each treatment with four replication. The observations were done at 12, 18, 24, 30 and 36 hours in an incubator at 37°C. Based on the data analysis, basil leaves (*Ocimum sanctum*) infusion has anthelmintic activity against *A. suum* *in vitro*. The greater of the concentration and the longer of time of immersion, will make the death percentage of *A. suum* become higher. Concentration of basil leaves (*Ocimum sanctum*) infusion 15% is the effective concentration that can kill 100% of the *A.suum* during 36 hours of immersion.

Key word: basil leaves, infusion, *Ascaris suum*, *in vitro*

Introduction

Ascaris suum is a parasite nematode that causes infection in swines with high prevalence rates in host populations. The prevalence of *A. suum* infection varies with geographical region and farm management practices but few swine herds are totally free of infection (Roepstorff, 2003). Swine ascariasis interferes with health and performance of swines while resulting in reduced feed gain ratios and liver condemnation incurring economic losses (Stewart and Hale, 1988). This parasite is usually associated with liver damages called “milk spots” caused by larvae migration, resulting in organ condemnation.

The usual treatment of ascariasis is done by using anthelmintic. Various problems have been found in the parasite control using synthetic

anthelmintic, chemical residues, toxicity issues, not economical and unavailability of these drugs in remote areas (Hussain, 2008). At present, ascariasis control is based mainly on mass treatment with synthetic anthelmintic drugs. In the long term this is not sustainable reinfection after annual or biannual drug treatment is more or less unavoidable due to the long lived and resistant eggs which survive for many years in the environment (Jia *et al.*, 2012). Moreover, the use of synthetic drugs is often not feasible for *A. suum* control in many swine production systems. Small holder farmers in many developing countries often do not have access to expensive anthelmintic drugs, many organic and low income farms are not able to prophylactically treat animals with synthetic drugs. Therefore, there is an urgent need

to investigate alternative or complementary options for the control of these parasites (Andrew *et al.*, 2014).

Indonesia rich in various kinds of medicinal plants and have been used for generations as a traditional medicine. One of the medicinal plants found in Indonesia is basil (*Ocimum sanctum*). Basil has been known from as the vedic period. Bhatt (2014) reported that basil extract has numerous pharmacological activities like hypoglycaemic, immunomodulatory, analgesic, antistress, antipyretic, antiulcerogenic, anti-inflammatory, antihypertensive and antibacterial. Recent work of its consumption has shown that basil had no genotoxic or organ toxic effects (Chandrasekaran *et al.*, 2013). Reported by Pandey and Madhuri (2010) different parts of basil are used in traditional medicine and cure of many illnesses and everyday ailments like common cold, headache, cough, flu, earache, fever, colic pain, sore throat, bronchitis, asthma, hepatic diseases, malaria fever, as an antidote for snake bite and scorpion sting, flatulence, migraine headaches, fatigue, skin diseases, wound, insomnia, arthritis, digestive disorders, night blindness, diarrhea and influenza.

Anthelmintic activity of basil leaves (*Ocimum sanctum*) against infections as it exhibited excellent prophylactic potential gastrointestinal nematodes has been evaluated by many authors. Joshi *et al.* (2013) reported that the aqueous extract of leaves of *Ocimum sanctum* showed good activity against *Pheretima posthuma*. Adult earthworms (*Pheretima posthuma*) were used due to their anatomical and physiological resemblance with the intestinal round worm parasite. Sentana (2010) also reported that ethanolic extract of basil leaves has anthelmintic activity against *A. suum* *in vitro*. As reported by Karumari *et al.* (2014), *Ocimum sanctum* phytochemical constituents contains flavonoid, phenol and tannin. Tannins and phenolics are known to interfere with the energy generation in helminth parasites by uncoupling oxidative phosphorylation and also bind to free proteins in the gastrointestinal tract of host animal or glycoprotein on the cuticle of the parasite and leading to death (Athnasiadou *et al.*, 2001).

In vitro model was used because of ethical considerations, and tend to find the closest model to *in vivo* research, beside that was to know the

chemical compound and to get first information of this herb for next modification *in vivo* research. Infusion dosage was used because it refers to pharmacopoeia standard for traditional medicine preparations. For comparison this research used piperazine citrate as the drug of choice of ascariasis treatment (Subekti *et al.*, 2012). Based on the background above this research wanted to determine the activity of basil leaves as anthelmintic against *A. suum* *in vitro* in the preparation of infusion.

Materials and Method

This research used completely randomized design to determine anthelmintic effect of basil leaves (*Ocimum sanctum*) infusion against *A. suum* *in vitro*. In this research, the volume of each treatment was 100 ml and contains 10 *A. suum* alive. The research observation started after 12, 18, 24, 30 hours of immersion and ended at 36 hours. This research was conducted in six treatments and four replication each treatment. P0:Immersion of *A. suum* into physiological NaCl 0.9% P1:Immersion of *A. suum* into aquadest, P2:Immersion of *A. suum* into piperazine citrate 1%, P3:Immersion of *A. suum* into basil leaves infusion 15%, P4:Immersion of *A. suum* into basil leaves infusion 25%, P5:Immersion of *A. suum* into basil leaves infusion 35%

Result and Discussion

Average of death percentage of *A. suum* within observation time, 12, 18, 24, 30 and 36 can be seen in Table 1.

Table 1 The Average of Death Percentage of *A. suum* within Observation Time .

Treatments	Death percentage during Observation Time (hours)				
	12	18	24	30	36
NaCl	0	0	0	0	0
Aquadest	0	0	2.5	20	70
PC1%	0	0	7.5	37.5	97.5
BLI 15%	7.5	15	47.5	90	100
BLI 25%	15	25	67.5	100	100
BLI35%	20	52.5	100	100	100

PC : piperazine citrate , BLI : basil leaves infusion

The average of death of *A. suum* due to treatment at 12, 18, 24, 30 and 36 hours can be seen in Table 2.

Table 2 The Average of Death of *A. suum* Due to Treatment Time

Treatment	The Average of Death of <i>A. suum</i> Due to Treatment Time (Hours)				
	12	18	24	30	36
NaCl	0.25 ^a ± 0.00	0.25 ^a ± 0.00	0.25 ^a ± 0.00	0.25 ^a ± 0.00	0.25 ^a ± 0.00
Aquadest	0.25 ^a ± 0.00	0.25 ^a ± 0.00	0.97 ^{ab} ± 1.45	4.39 ^b ± 0.94	8.35 ^b ± 0.48
PC1%	0.25 ^a ± 0.00	0.25 ^a ± 0.00	2.43 ^b ± 1.45	6.11 ^{bc} ± 0.42	9.84 ^c ± 0.24
BLI 15%	2.43 ^b ± 1.45	3.34 ^b ± 2.26	6.72 ^c ± 1.72	7.77 ^c ± 3.08	9.96 ^c ± 0.00
BLI 25%	3.48 ^c ± 0.65	4.93 ^c ± 0.92	8.21 ^c ± 0.31	9.96 ^d ± 0.00	9.96 ^c ± 0.00
BLI 35%	4.47 ^d ± 0.00	7.23 ^d ± 0.33	9.96 ^d ± 0.00	9.96 ^d ± 0.00	9.96 ^c ± 0.00

^{a-d} Different superscript in the same column indicate significant difference (p<0.05)

From the results of research, after 12 hours the death percentage of *A.suum* found in basil leaves infusion 15% was 7.5%, in basil leaves infusion 25% was 25% and in basil leaves infusion 35% was 20%. At the 18 hours after treatment, the death percentage of *A. suum* in basil leaves infusion 15% was 15%, basil leaves infusion 25% was 25%, and in basil leaves infusion 35% was 52.5%, while the other treatments still not obtained the death of *A. suum*. Subsequent observations conducted at 24, 30 and 36 hours. At the 36 hours obtained 100% of *A. suum* were dead at all concentrations of basil leaves infusion (15%, 25% and 35%) and in piperazine citrate 1% was 97.5%, whereas in NaCl was still no death *A. suum*, and in aquadest the death percentage of *A. suum* was 70%. The result of the death percentage during observation time 12, 18, 24, 30 and 36 can be seen in Table 1.

From the data analysis obtained, NaCl and aquadest showed significantly different among the three concentrations of basil leaves infusion. The higher concentration infusion of basil leaves the faster *A. suum* will die, besides that the death time of worm in immersion of basil leaves infusion faster than in aquadest immersion, so it can be concluded that death was due to the influence of the treatment. Concentration of basil leaves infusion 15% was the best concentration to kill *A. suum* within 36 hours compared to 1% of piperazine citrate and other basil leaves infusions. The result can be seen in Table 6.

The results of this study showed that basil leaves infusion have anthelmintic activity against *A. suum in vitro*. Allegedly basil has potential as anthelmintic because of the content of chemical elements phenolics, flavonoid, tannins and saponins, it consequent with previous research,

Ocimum sanctum has phytochemical constituents contains flavonoid, phenol and tannins (Karumari *et al.*, 2014). Tannins and phenolics are known to interfere with the energy generation in helminth parasites by uncoupling oxidative phosphorylation and also bind to free proteins in the gastrointestinal tract of host animal or glycoprotein on the cuticle of the parasite and leading to death (Athnasiadou *et al.*, 2001). Anthelmintic activity of basil leaves was from the active substance content of saponins and tannins (Sentana, 2010). This research approve that basil leaves infusion has anthelmintic activity against *A. suum in vitro*.

Conclusion

From the results of research concluded infusion of basil leaves had anthelmintic activity against *A. suum in vitro*. The higher of concentration of basil leaves infusion given higher death percentage of *A. suum*, there was a possibility caused by active chemical substance that acts as anthelmintics.

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