

## Efficacy of Polyherbal Oil Against Gastrointestinal Nematode Worms in Kintamani Dogs

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**Abstract.** Gastrointestinal helminth in animals can be zoonotic-parasitic. This study aimed to determine the efficacy of polyherbal oil formulations against intestinal helminth parasites of Kintamani puppies. This study used 24 Kintamani puppies aged 3-6 months which were reared by means of release. The anthelmintic potency of polyherbal oil formulations was determined by measuring Eggs Per Gram of feces (EPG), using the Modified Stoll Dilution method before and seven days after treatment. The results showed that there was a decrease in the EPG value sequentially P0, P1, P2, and P3 that was -4%, 23.1%, 66.7%, and 81.3%. Based on the identification of helminth eggs, it was shown that the efficacy of polyherbal oil against *Toxocara canis* decreased EPG at P1, P2, and P3 by 26.09%, 73.33%, and 86.08%. Against *Ancylostomata* spp sequentially at P1, P2 and P3 caused a decrease in EPG of 17.39%, 52.63%, and 72.0%. Against *Strongyl* spp, respectively at P1, P2 and P3 caused a decrease in EPG by 16.67%, 58.82%, and 73.68%. Statistical analysis showed that polyherbal oil had a significant effect ( $P < 0.05$ ) on the decrease in EPG. It can be concluded that the polyherbal oil formulation has vermifugal efficacy against intestinal helminth parasites in Kintamani puppies.

**Key words:** Kintamani dog, Efficacy, Helminth Parasitic, Polyherbal Oil

### I. INTRODUCTION

The existence of dogs since time immemorial is very closely related to human life as house guards, hunting dogs, shepherd

dogs, and also because of the myth of dogs that bring good luck. The increasing socioeconomic status of the community has encouraged them to have their pet, one of

which is the popular dog. Dogs are pets that are very loyal to their owners [1], as loyal friends when lonely and also to reduce stress for their owners [2][3]. The Kintamani dog is germplasm with an attractive appearance, medium in size, and smart to be trained to be one of the prima donnas of pet dogs. The increasing number of Kintamani dog hobbyists has an impact on increasing demand and the dog population. A dense population without a balance of environmental carrying capacity will be at risk of disease that can be caused by viruses, bacteria, and parasites [4].

Parasitic disease in dogs can be caused by endoparasite and ectoparasite infestation. Endoparasite diseases in dogs caused by helminth include Ascariasis, Dirofilaria, Toxocariasis, and Ankylostomiasis [5][6]. There are parasitic diseases in dogs that are zoonotic parasitic [7][8]. This disease is also found to occur in Kintamani dogs, but very little has been published. In general, young animals are more susceptible to parasitic infestations than adult dogs. The incidence of nematode helminth infection in the gastrointestinal tract of Kintamani dogs was reported by [9][10].

Efforts to prevent and treat diseases in dogs start with the cleanliness of animals and

their cages, providing good nutrition, vaccination, treatment, and control of sick animals by consulting a veterinarian [11]. Veterinary medical action for endoparasitic helminthiasis cases can be done by administering deworming medication based on the diagnosis [12]. Nowadays, the use of herbal medicines to maintain health and treatment for pets is increasing [13]. This is based on the belief that natural herbal medicines are safer and a cultural heritage. For people who are difficult to reach for veterinary medical services, natural veterinary medicines can still be pursued on their own [14]. Various herbal plants such as bangle, ginger, galangal, turmeric, temulawak, weeds, coconut, and betel tree were reported to be effective in overcoming parasitic diseases in animals both in vitro and in vivo. Ginger is reported to be effective as antitoxoplasmosis [15]. In vitro neem leaves as anti-ectoparasitic in *Rhipicephalus sanguineus* and *Heterodoxus* spp. [16]. The herbs turmeric, temulawak, bangle, and betel leaf are reported to be effective as antiparasitic in vitro and in vivo [17] [18] [19]. Based on the above background, a study was conducted to determine the vermifugal effect of a polyherbal oil formulation consisting of a variety of local herbal

Simplicia with coconut oil (*Oleum cocos*) as a vehicle in Kintamani puppies.

## II. MATERIAL AND METHODS

### The Animals and Locate of Research

The object of the research was 24 Kintamani puppies aged 3-6 months which were kept extensively. The research location is in Sukawana Village, Kintamani District, Bangli Regency. Manufacture of polyherbal oil at the Veterinary Pharmacy Laboratory, Faculty of Veterinary Medicine, Udayana University. Examination of dog feces samples was carried out at the Denpasar Veterinary Center, Bali.

### Polyherbal Oil Formulation

Polyherbal oil is formulated from various herbal simplicial with medicinal properties, namely bangle, ginger, galangal, turmeric, temulawak, reeds, betel leaf, and coconut oil. Each simplicia was determined as much as 2% with coconut oil as the vehicle medium and with fermentation technology. The polyherbal oil was then weighed to determine the weight of the dosage per milliliter (mg/ml). The preparations obtained were stored in bottles and were ready to be used in research to see their potential as a gastrointestinal deworming in Kintamani puppies.

### Anthelmintic Potential of Polyherbal Oils

This study used a completely randomized design. To see the anthelmintic potential of polyherbal oil, 24 Kintamani puppies aged 3-6 months that were positively infected with gastrointestinal nematode helminth were randomly divided into four groups. The P0 group as control was given a placebo, which was distilled water. The treatment groups P1, P2, and P3 were given polyherbal oil at doses of 300, 600, and 900 mg/kg body weight for three days. The dose used is the development of the research method published by [20]. A sampling of feces was carried out in the morning using a stool pot, before and on the seventh day after the first treatment. Examination of stool samples using the Modified Stoll Dilution method [21], to determine the EPG. The anthelmintic potential was seen from the decrease in EPG after treatment with polyherbal oil doses. The working steps of the Modified Stoll Dilution method are as follows three grams of feces are taken in a test tube (45 ml. graduated). Fill the tube up to 45 ml. with N/10 NaOH (4 gms of NaOH, 1 lit. of distilled water) and add 10-12 glass

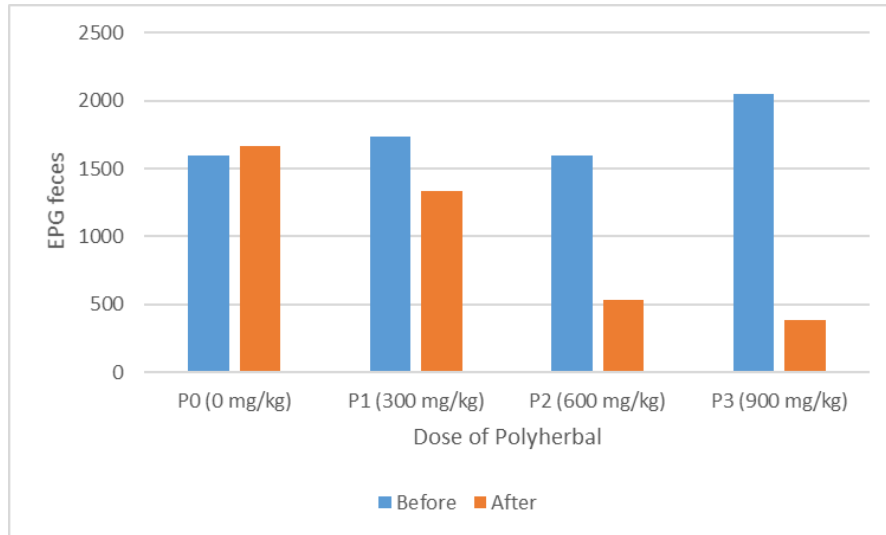
beads. Close it with a stopper and homogenize the fecal material, and 0.15 ml of the suspension is drawn with a pipette and placed on a slide and a coverslip is applied. Count the total number of eggs. Multiply the number of eggs by 100, which gives the eggs per 1 gram of feces.

### **Data Analysis**

The research data in the form of EPG will be presented descriptively in the form of mean and standard deviation. Data analysis used the ANOVA test, and if there was a significant difference ( $P < 0.05$ ) between the treatment groups, it was continued with Duncan's test (Ali and Bhaskar, 2016). The data of Eggs Per Gram (EPG) will be presented in Mean ( $\pm$ SD) and analyzed using One Way Analysis of Variance followed by Duncan's test.

### **III. RESULT**

The results showed that the polyherbal oil formulation was able to significantly reduce the EPG of gastrointestinal worms in Kintamani puppies ( $P < 0.05$ ) compared to the control group. The intensity of helminth infection is reflected by the magnitude of the EPG. The mean EPG before and after treatment in the groups P0:  $1600 \pm 283$  and  $1667 \pm 234$ , P1:  $1733 \pm 383$  and  $1333 \pm 197$ , P2:  $1600 \pm 358$  and  $533 \pm 82$ , and P3:  $2050 \pm 345$  and  $383 \pm 133$ . The highest decrease in EPG occurred in the P3 group (900 mg/kg dose) at 81.3% and followed by the P2 group by 66.7% and P1 by 23.1%. Meanwhile, in the control, there was an increase in EPG of 4%. In detail, the potential efficacy of polyherbal oils is presented in Figure 1 and Tables 1, 2, and 3.



**Figure 1.** The average EPG of Kintamani puppies feces given polyherbal oil

**Table 1. Eggs Per Gram *Toxocara canis* after administration of polyherbal oil**

Group	N	Mean of EPG				
		Initial	SD	Latter	SD	Diff (%)
P0	6	1.017	214	1.050	52	3,28
P1	6	1.150	259	850	55	(26,09)
P2	6	1.000	219	267	82	(73,33)
P3	6	1.317	271	183	17	(86,08)

**Table 2. EPG *Ancylostoma* spp. after administration of polyherbal oil**

Group	N	Mena of EPG				
		Initial	SD	Latter	SD	Diff (%)
P0	6	350	105	367	82	4,76
P1	6	383	147	317	33	(17,39)
P2	6	317	147	150	84	(52,63)
P3	6	417	117	117	41	(72,00)

**Table 3. EPG Strongyl spp. after administration of polyherbal oil**

Group	N	Mean of EPG				
		Initial	SD	Latter	SD	Diff (%)
P0	6	233	103	250	55	7,14
P1	6	200	89	167	52	(16,67)
P2	6	283	75	117	41	(58,82)
P3	6	317	75	83	41	(73,68)

#### IV. DISCUSSION

Examination of 24 samples of Kintamani dog feces aged 3-6 months that were kept extensively in Sukawana Village, Kintamani, Bangli Regency, Bali was found to be positive for helminthiasis infection. This is reinforced by the owner's confession that he has never been given deworming medicine. Parasitic disease in the digestive tract of dogs is dominated by nematode worms. Cases of nematodiasis can cause symptoms such as weakness, lethargy, dull hair, and thinness [21]. Several studies revealed cases of helminthiasis in Kintamani Dogs in their natural habitat, including Ancylostomiasis with a prevalence of 55.55% [9], and Toxocariosis reaching 22.22% where infestations in young dogs were higher than adults but not significant [10]. Meanwhile, reports on cestode helminth infestations in Kintamani dogs are still very

limited. [22], reported the average prevalence of *Dypilidium caninum* of 18% and the prevalence of *Taenia* spp. by 10% in Balinese dogs in Denpasar City. Helminthiasis needs serious attention because they are parasitic-zoonotic [6]. Co-infection with nematodiosis and coccidiosis has been reported to cause severe enteritis in the host [23].

In this study, identification of helminth eggs showed that the cause of helminthiasis in Kintamani dogs was *Toxocara canis*, *Ancylostoma* spp, and *Strongyl* spp. To determine the level of worm infestation, the total eggs per gram (EPG) of feces were calculated. The P0 group as control was given a placebo. Groups P1, P2 and P3 were given Polyherbal oil at doses of 300, 600, and 900 mg/kg body weight for three consecutive days. The results showed that there was a total decrease in EPG sequentially P0, P1, P2, and P3 which were -4.17%, 23.08%, 66.67%,

and 81.30%. Based on the identification of helminth eggs, it is known that the efficacy of polyherbal oil on *Toxocara Canis* EPG sequentially at P1, P2, and P3 caused a decrease in EPG values of 26.09%, 73.33%, and 86.08%. The EPG of *Ancylostomata* spp respectively at P1, P2 and P3 caused a decrease of 17.39%, 52.63%, and 72%. Analysis of variance showed that the polyherbal oil treatment had a significant effect on reducing the EPG of dog gastrointestinal helminth compared to controls. A decrease in EPG indicates the number of helminths laying eggs is decreasing or the worms are dying. Treatment was declared effective if the vermifugal power was at least 72%.

Previous studies reported that secondary metabolites such as phenylbutanoid compounds, alkaloids, flavonoids, curcumin, saponins, tannins, steroids, and terpenoids have pharmacological activity as anthelmintics. Secondary metabolites can act singly, additively, or synergistically in causing paralysis (paralysis) and/or causing the death of helminths. These active compounds have a unique mode of action and can act at one or several target sites on the helminth's body. The pure extract of the bangle (*Zingiber*

*purpureum*) which contains flavonoids, alkaloids, and saponins at a concentration of 16% effectively kills *Ascaris suum*.

Flavonoids have low toxicity to animal cells, however, several types of flavonoids such as genistein, kaempferol, rutin, quercetin have shown damaging effects on helminths. These flavonoid compounds can inhibit specific enzymes, stimulate hormones and neurotransmitters in the helminth's body. This causes to block the response of the worm's muscles to the accumulation of acetylcholine. Excessive stimulation has an impact on depression in the nervous system followed by convulsions, paralysis, and death of the helminth. Flavonoids are rapidly absorbed and cause denaturation of the intestinal tract proteins of the helminths, thereby disrupting the growth of filarial or young helminths. [24]. The mechanism of action is similar to that of piperazine citrate pharmacodynamics which causes paralysis and death of helminths.

The alkaloids and glycosides have been reported to be very effective as an anthelmintic [25]. However, its role as an anthelmintic is not known, whether it acts as ovicidal, larvacidal, and vermifugal. According to research by [26] alkaloids, such as arecholine and tannins, have anthelmintic

effects. Arecholine is toxic and causes paralysis. Tannins can inhibit enzymes and damage membranes. The membrane damaged by tannins causes paralysis which eventually leads to the death of the embryo. Tannins have the ability to precipitate free proteins in the helminth's gastrointestinal tract or glycoproteins in the helminth cuticles, thereby disrupting physiological functions such as motility, absorption, and reproduction. Tannins also work to disrupt the helminth's energy formation process by breaking oxidative phosphorylation which causes the worms to weaken or die [27].

Saponins can lower the surface tension of aqueous solutions so that the contact between the infusion with the helminth skin becomes faster and more effective. In addition, saponins can irritate mucous membranes. Saponins are known to cause  
 12 food refusal and starvation so that the helminths will lack energy and die [28].

## V. CONCLUSION

The polyherbal oil formulation was able to significantly reduce EPG. The best results were shown at a dose of 900 mg/kg body weight, which was able to reduce EPG up to 81.3%. Therefore, we conclude that the polyherbal oil formulation has vermifugal

efficacy against gastrointestinal helminth parasites of Kintamani puppies. In the future, further research is needed to obtain the most effective doses of vermifugal, larvicidal, and ovicidal.

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