

The Effect of *Lentinus squarrosulus*, (Mont.) Consumption on the treatment of Gastritis in Male Wistar Rats

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ABSTRACT

Introduction: Gastritis or /and gastric ulcers are one of the most common symptoms which occur in a large amount of the population. It results in a very high amount of medication expense. This study investigated the prevention and treatment of gastritis and /and gastric ulcer actions using white log mushroom (*Lentinus squarrosulus*, (Mont.). **Methods:** White wistar rats were induced with 100 % absolute alcohol to be effective for gastritis and /or ulceration within rats. The gastric lesions of white wistar rats were investigated and calculated the lesions of stomach areas under microscopic method. The results were calculated and a comparative study among white wistar rats. Ulcer prevention and ulcer healing properties were administrated though a dose of 250 mg/kg of *L. squarrosulus* extract and *L. squarrosulus* compared with 50 mg/kg of Ranetidine. **Results:** It was found that *Lentinus squarrosulus*, (Mont.) consisted of Beta-glucan was the active ingredient creating a healing effect. It showed a result of 100% healing effect in gastritis and/or ulcers in which it had similar healing effects between extracted solutions of white log mushroom (250 mg/kg body wt.) The healing activity also showed the equivalent effect compared to Ranetidine 150mg. **Conclusions:** Extracted solution of white log mushroom showed prevention and treatment efficacy in gastritis and/or ulcer are equivalent to Ranetidine 150 mg. A similar dose of 250 mg/kg of *L. squarrosulus* extract and *L. squarrosulus* snack can prevent and heal ulcers and is more effective than Ranetidine.

Key words: *L. squarrosulus*, Ulcer prevention, Ulcer healing, Beta glucan, Ranetidine 150 mg.

INTRODUCTION

Mushrooms have highly nutritious compounds, which are used to prevent and treat disease and the ingredients can be used in supplements. Especially, peptic ulcer or gastritis disease occurring from the infection of *Helicobacter pylori* as a Gram-negative bacteria in the human stomach¹⁻⁶ and administration of Non-steroidal Anti-inflammatory drugs (NSAIDs). These drugs are used for the treatment of pain, rheumatic, and cardiovascular diseases. Regular use of NSAIDs will inhibit an action of Prostaglandins which can affect the digestive system, liver, cardiovascular system and respiratory system and also the central nervous system.⁷⁻¹⁰ found that there are increasing numbers of gastritis and ulcer incidences throughout the world.

White log mushroom has high nutritional values and no fat which are essentials for human body such as protein, carbohydrate, selenium, iron, magnesium, potassium, zinc, manganese, vitamin, vitamin B1, vitamin B2, vitamin.^{3,7} The contents of white log mushroom consist of twice the protein than vegetable leaves and or an oat diet⁹ including higher potassium in order to decrease in anti-hypertension.^{9,7,4} mentioned that it has high antioxidant levels which can have a healing effect in preventing and curing the gastric ulcer and also^{8,7} found an increase of human immunity and longevity (Thailand Department of Traditional and Alternative Medicine Ministry of Public Health

2013). Active ingredients in white log mushroom are phenolic, tannin and flavonoids,^{7,4} polysaccharide, and triterpenoid.etc. Reported that mushroom in *Lentinus* family contain high amounts of Beta glucan which is called lentinans. Lentinans has an anticancer property and is a high immunity modulator and also has high antioxidant activity which are confirmed by DPPH Assay analysis.¹¹ In 2011, we found that white log mushroom can give the prevention of gastritis or ulcer action. The hot water extraction of white log mushroom solution contained protein (57.6g/100 g) carbohydrate, selenium, iron, magnesium, potassium, zinc, manganese, vitamin A, vitamin E, Vitamin B1, vitamin B2 vitamin B3.

MATERIALS AND METHODS

Study area

The experiment was carried out in November 2018 to March 2019 at animal laboratory faculty of sciences, Khon khan University

Preparation of *L. squarrosulus* extract

L. squarrosulus was obtained from mushroom farm at Ban Dong, Wongong Sub-district, Muang Mahasarakham, Maha Sarakham. *L. squarrosulus* samples were extracted by water at a ratio of 1: 1 and boiled for 30 minutes. The broth was centrifuged at 3000 g for 15 minutes and the supernatant was filtered using Whatman no. 1 filter paper. The water extract was freeze dried.

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Preparation of *L. squarrosulus* (Mont.) snack

White log mushroom 10 kg were washed with purified water and were processed using a freeze drying machine for 4-5 hours. The freeze dried white log mushroom was grinded and mixed with starch and flavouring powder to taste. The products were made into layered sheets and were cut into thin pieces and put into an oven in order to make them crispy. They were kept in sealed containers awaiting further testing.

Preparation of wistar rat subjects

Adult male wistar rats aged 6–8 weeks and weighed between 180 and 200 g were purchased from Northeast Laboratory Animal Centre, Khon Kaen University, Thailand. The animals were housed at $27 \pm 2^\circ\text{C}$ temperature, fed with standard laboratory pellets, and provided with water ad libitum.

Ulcer prevention property

A total of 24 of SD rats were divided randomly into four groups comprised of six rats in each group. All groups were deprived of food for 24 hours before the experiment. The experiment began with pretreatments according to the assigned group:

Group 1 (ulcer control) received the vehicle (distilled water) only

Groups 2 received *L. squarrosulus* extract 250 mg/kg

Groups 3 received *L. squarrosulus* snack 250 mg/kg

Groups 4 received (positive control) 50 mg/kg of ranitidine 150 mg.

All wistar rat subjects were administered with absolute ethanol after thirty minutes of the pretreatment. After an additional thirty minutes, all subjects were examined and their stomachs were removed and kept immersed in 10% of buffered formalin before the analysis of gastric lesions.

Ulcer healing property

A total of 24 SD rats were divided randomly into four groups comprising six rats in each group:

Group 1 animals which served as normal control received vehicle (distilled water) only.

Groups 2, 3 and 4 (ulcerated groups) were pre fasted for 24 hours before inducing ulcer using absolute ethanol (5 mL/kg).

Group 2 was treated with *L. squarrosulus* extract at a dose of 250 mg/kg

Group 3 was treated with *L. squarrosulus* snack at a dose of 250 mg/kg

Group 4 was treated with 50 mg/kg ranitidine 150mg (positive control).

All groups were treated once daily in the morning. One rat per group was examined after a 24 hour treatment. The stomach tissues were homogenized with phosphate buffer saline (PBS). Both serum and tissue homogenates were stored at -80°C for analysis.

Gross evaluation of gastric lesions

Each stomach was incised along a greater curvature and rinsed with distilled water to remove gastric contents. The stomach was examined under a dissecting microscope (1.8x) with a square grid eyepiece. (Big square: length \times width = $10 \times 10\text{mm}^2$ = ulcer area) to access the formation of ulcerated area (hemorrhagic lesions). The sum of all lesions, in mm^2 , for each stomach was expressed as the ulcer area (mm^2). The percentage of inhibition (%) was calculated by the following formula Omar:

$$\% \text{ inhibition is equal to } \frac{\text{UA}_{\text{control}} - \text{UA}_{\text{treated}}}{\text{UA}_{\text{control}}} \times 100$$

Statistical analysis

The results were expressed as mean \pm S.E.M. Statistical differences among groups were evaluated by Dunnett's multiple comparison test. Student's t-test was applied to comparisons between two groups. P values of $<.05$ were considered significant.

RESULTS AND DISCUSSION

Prevention of ulcer

The result showed *L. squarrosulus* extract and *L. squarrosulus* snack reduced the formation of gastric ulcers induced by ethanol and can inhibit more than ranitidine 150 mg. The inhibition percentage of ulcer area was 100%, 88%, and 84% respectively (Table 1, Figure 1).

Healing of ulcer

After administration of 250 mg/kg of *L. squarrosulus* extract and *L. squarrosulus* snack for 48 hours increases the healing of gastric ulcer in ethanol-induced rats. This study found that *L. squarrosulus* extract, *L. squarrosulus* snack and ranitidine 150mg can decrease the ulcer area by 100%, 86%, and 81% respectively (Table 2).

The comparative of efficacy of *L. squarrosulus* White Log Mushroom and White Log Mushroom Snack and Ranitidine 150 mg. in prevention and treatment of gastric ulcers and gastritis in wistar rats found that induced absolute alcohol wistar rats had the result of 81 – 100 % healing effect of gastritis and ulcer which was equal to ranitidine 150 mg. Other studies showed that Beta glucan in white log mushroom had an inhibition action of histamine secretion from gastric cells which can decrease the secretion of gastric acid or HCL.⁷ It was experimented in 125, 125, 250 and 500 mg/kg of white mushroom extraction administered to wistar rats found that the healing effect of induced absolute alcohol ulcer wistar rats have equivalent activity with Ranitidine 150 mg efficacy. The maximum healing result was found in using dosage of 250 mg/kg white mushroom extraction administered to wistar rats which had positive efficacy equivalent to ranitidine 150 mg.

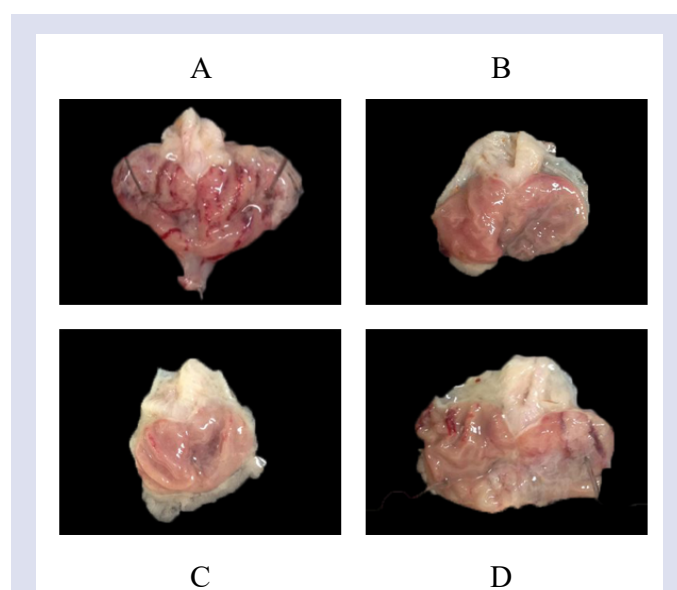


Figure 1: Show wistar rats with induced absolute ethanol gastritis (A = control wistar rats) B = wistar rats with administered white log mushroom extraction 250 mg/kg C = wistar rats with administered white log mushroom extraction 250 mg/kg from white mushroom snack D = wistar rats with administered ranitidine 150 mg at dosage of 50 mg/kg body weight.

Table 1: Prevention of gastric ulcer (Mean \pm SEM; n=6).

Treatment	Ulcer area (mm ²)	% Inhibition
Distilled water	181.90 \pm 13.38 ^c	-
<i>L. squarrosulus</i> extract at dose 250 mg/kg	-	100
<i>L. squarrosulus</i> snack at dose 250 mg/kg	20.47 \pm 6.89 ^{ab}	88
ranitidine 150mg at dose 50 mg/kg	28.85 \pm 4.14 ^b	84

All values were expressed as mean and \pm standard error mean of six replicates of wistar rats. Means with different superscripts was significantly different ($P < .05$).

Table 2: Healing of Ulcer of gastric ulcer (Mean SEM; n=6).

Treatment	Ulcer area (mm ²)	% Inhibition
Distilled water	135.72 \pm 3.72 ^b	-
<i>L. squarrosulus</i> extract at dose 250 mg/kg	-	100
<i>L. squarrosulus</i> snack at dose 250 mg/kg	17.68 \pm 12.20 ^a	86
ranitidine 150mg at dose 50 mg/kg	25.55 \pm 16.44 ^a	81

Also, It has been shown that mushroom extracts revealed similar electrochemical responses, suggesting similar electroactive chemical composition, and oxidation potentials more positive than those of the standards (ascorbic and gallic acids).² Some research has been reviewed in case of antidiabetic property of mushrooms. Therefore, it is proposed further research of functional mushrooms for preventive and curative measures for diabetes and its complications.⁵ This study demonstrated that both a dose 250 mg/kg. Of *L. squarrosulus* extract and *L. squarrosulus* snack can prevent and healing ulcers and more effective than ranitidine 150 mg.

SIGNIFICANCE STATEMENT

My findings have revealed that there is an active ingredient named Beta Glucan in White log mushroom (*Lentinus squarrosulus*) which can produce the efficacy of healing effect in gastro intestinal tract infection. It can reduce gastritis and similar results of actions using Ranitidine 150 mg. in Male Wistar Rats.

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AUTHORS' CONTRIBUTION

Experimental Areas referring to comparative ulceration healing efficacy between White log mushroom and Ranitidine 150 mg.

CONFLICTS OF INTEREST

None.

LIVEDNA

66.14375*.

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Working Experience

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