Utilization of Bark Condensed Tannin as Natural Preservatives Against Subterranean Termite

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**Abstract.** Extractive-based preservative has been isolated from many parts of plant to protect wood from fungi or pest. This is due to the presence of bioactive chemical as a natural preservative. Present study, the condensed tannin was isolated from bark-waste of *Acacia crassicarpa* and *Acacia mearnsii*. The yield of the extracts obtained from *A. crassicarpa* using a 70% acetone aqueous solution (7% based on bark weight) is less than that obtained from *A. mearnsii* (34%). To evaluate the termiticidal activity of condensed tannin from both extracts, no-choice tests were conducted using *Reticulitermes speratus*. After 14 days, mass loss of the dimmer condensed tannin treated was less than 50%, significantly different with control. In addition, a monomer condensed tannin, catechin was presented the similar result. Meanwhile, the termite mortality of catechin is slightly higher than the dimmer. According to both parameter, the medium level of mass loss and termite mortality, the dimmer suggested as antifeedant in mechanism of termiticidal. The feeding activity is decreased after contact with the treated paper disc. In polymer condensed tannin, the result seems similar to dimmer condensed tannin. Condensed tannin and hydrolyzable tannin which were contained in the bark effected termite consumption because it caused inactivation of enzyme in termite digestion system. In fact, many factors effect termite activity from natural extractives such as compound type, quantity and also extract retention activity. The finding of this study should contribute to the developing of natural wood preservative from extractive-based. Especially, the study of structure-active relationship (SAR) of condensed tannin from *A. crassicarpa*, which can improve the utilization of bark-waste from pulp industry.

1. **Introduction**

Natural wood preservative based on plant extracts, have been known to protect wood from termite. Many studies have been reported about the effectiveness of reduced chemical or even chemical-free treatments which was compared to the use of plant extracts against termite [1,2]. Due to the effect of synthetic or chemical pesticide into the environment, the use of bio-termiticide has been increase although in still limited. The natural preservative based on plant extracts can be isolated from any part of plant such as heartwood [3,4], bark [5,6], leaves, fruit and seed [7,8].
In other situation, bark of *A. crassicarpa*, by-product of the pulp industry as a source of extractive has remains unutilized. Indonesia as the third largest manufacturer of pulp and paper in Asia, it is produces a significant amount of bark by-products. Bark contains a high concentration of tannin; as a result, interest in developing methods for utilizing tannin has recently increased. Unfortunately, there is no information that reported about studies on tannin from the bark of *A. crassicarpa*. Pietarinen et al. 2005 only reported about the polyphenolics compounds from the knot and stem wood of *A. crassicarpa* [9].

So, this study aims to utilize the bark waste of *A. crassicarpa* as natural preservative towards subterranean termite

2. Material and Methods

2.1. Isolation of condensed tannin from bark of *A. crassicarpa*

Bark of *A. crassicarpa* was collected from 4-5 year-old trees grown on Research and Development plantation area of PT. Riau Andalan Pulp and Paper (PT. RAPP) located in Pelalawan area, Riau, Indonesia at 2011. The isolation of condensed tannins were conducted according to procedure that reported in Ismayati, et al., 2017[10]

2.2. No-choice feeding tests against *Reticulitermes speratus* using condensed tannin samples from *A. crassicarpa* and *A. mearnsii*

*R. speratus* was collected from the Living Sphere Simulation Field, Research Institute for Sustainable Humanosphere (RISH), Kyoto University (Kagoshima Prefecture, Japan) and maintained at the University of Tsukuba (Ibaraki Prefecture, Japan), under controlled laboratory conditions (28°C and 80% relative humidity).

The condensed tannin sample (C-EA-1, C-WS-AW, and M-WS-AW) was applied into no-choice feeding test according to previous report [10,11]. As a control, untreated paper discs, solvents treatment and catechin were evaluated in the same manner as the discs with the extractives in this no-choice feeding test as the control.

To measure termiticidal activity, the mass loss of paper disc was calculated at the end of test periods and the dead termites were also counted. Three replicates were performed for each sample.

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\text{Mass loss} \% = \frac{\text{ODS}_1 - \text{ODS}_2}{\text{ODS}_1} \times 100\% 
\]

\[
\text{Termite Mortality} = \frac{\text{Number of died termite}}{50} \times 100\% 
\]

2.3. Statistical analysis

The effects condensed tannin on mass loss and termite mortality were examined using an analysis of variance (ANOVA), and samples showing significant differences (P < 0.05) were analyzed using Duncan’s post hoc test (\(\alpha = 0.05\)). All analyses were performed using IBM SPSS version 22 (IBM Japan, Tokyo, Japan).

*Figure 1. A. crassicarpa* tree planted at PT. RAPP, Indonesia (a), bark sample of *A. crassicarpa* (b) and No-choice feeding test using *R. speratus* (c)
3. Result and Discussion

The results of the no-choice feeding test to evaluate the effects of condensed tannin treatments are shown in Figure 3. After 14 days, mass loss of condensed tannin treatments (C-EA1, C-WS-AW, and M-WS-AW) were less than 50%, significantly different with treatment control. As dimmer control, catechin was used as control that presented the similar result. As second parameter, the termite mortality of catechin is slightly higher than C-EA1. Based on those result, C-EA1 shows a medium level as natural wood preservative, which suggested as antifeedant in mechanism of termiticidal. After contact with extract, the feeding activity is decreased [12].

A similar result shown by polymer condensed tannin as well as in dimmer treatments. Both polymer condensed tannin from A. crassicarpa and A. mearnsii (C-WS-AW and M-WS-AW) have a same trend of mass loss and termite mortality in the medium level. Based on these result, it was suggested that polymer condensed tannin can caused by the decreasing of feeding activity of termite due to the inactivation of enzyme in the termite digestion system [13,14]

![Figure 2](image)

**Figure 2.** Box plot of mass loss and mortality of R. speratus in no-choice feeding tests in Tectona grandis crude extractives and MAQ.

Legends: C-EA1= Dimmer condensed tannin from A. crassicarpa; C-WS-AW = Polymer condensed tannin from A. crassicarpa from; M-WS-AW = Polymer of condensed tannin from A. mearnsii

Bio-assay activity of compounds are related to their chemical structures. The structure-active relationship of C-EA1 (flavan-3-ols) that lacks an oxygen at C3 of pyran ring is very interesting for further study. To identify which functional group related into the bioassay activity, the C-EA1 is compared with Quercetin and Fisetin. Previous study, Quercetin that has a hydroxyl group in the C-5 A-ring showed higher antifeedant activity than Fisetin [15]. C-5 and C-7 hydroxyl groups in A ring of Quercetin showed higher antifeedant than Fisetin which do not have C-5 This indicates that the C-5 and C-7 hydroxyl groups in the A-ring are important for anti-termite activity [10]. The dimmer of C-EA1 that lacks an oxygen at C3 of pyran ring is very interesting for further study.
4. Conclusion
The dimmer condensed tannin C-EA1 and polymer condensed tannin from *A. crassicarpa* and *A. mearnsii* are potentially useful as a termite control agent. They act as antifeedant, against *R. speratus*.

5. References
[1] Kim YS, Singh AP, Wong AHH, Eom TJ, Lee KH 2006 Micromorphological and chemical characteristics of cengal (*Neobalanocarpus heimii*) heartwood decayed by soft rot fungi. *Mokchae Konghak* 34(2), 68-77
[2] Rodrigues AMS, Amusant N, Beaucene J, Eparvier V, Lemenager N, Baudass C, Espindola LS, Stien D 2011 The termitecidal activity of Sextonia rubra (Mez) van der Welf Lauraceae) extract and its active constituent rubrynomide *Pest Manag Sci* 67(11): 1420-1423
[3] Chen K, Ohmura W, Doi K, Aoyama M 2004 Termite feeding deterrent from Japanese larch wood. *Bioresource Technology* 95: 129-134
[4] Ohmura W, Doi S, Aoyama M, Ohara S 2000 Antifeedant activity of flavonoids and related compounds against the subteranean termite Coptotermes formosanus Shiraki. *J Wood Sci* 46: 149-153
[5] Viviers PM, Botha JJ, Ferreira D, Roux DG, Saayman HM 1983 Synthesis of condensed tannins. Part 7. Angular [4,6;4,8]-prorobinetinin triflavonoids from black wattle (‘Mimosa’) bark extract. *J Chem Soc Perkins Trans* 1: 17-22
[6] Yazaki Y, Collins PJ 1994 Wood adhesives from Pinus radiata barks of some pine and spruce species. *Holz als Roh-und Werkstoff* 52: 185-190
[7] Tarmadi D, Setiawan KH, Yusuf S 2004 The Efficacy of the Oleic Acid Isolated from *Cerbera Manghas* L. Seed Against a Subterranean Termite, Coptotermes gestroi Wasmann and a Drywood Termite, *Cryptotermin s cynocephalus* Light. *Procedia Environmental Sciences* 20: 772-777
[8] Setiawan KH, Tarmadi T, Ismayati I, Yusuf S 2013 Bioefficacy Performance of Neem-based Formulation on Wood Protection and Soil Barrier against Subterranean Termite, *Coptotermes gestroi* Wasmann (Isoptera: Rhinotermitidae). *Procedia Environmental Sciences* 17: 135-141
[9] Pietarinen SP, Willfor SM, Sjoholm RE, Holmbom BR 2005 Bioactive phenolic substances in important tree species. Part 3: knots and stemwood of *Acacia crassicarpa* and *A. mangium*. *Holzforschung* 59:94–101
[10] Ismayati M, Nakagawa-izumi, A, Ohi H 2017 Structural elucidation of condensed tannin from the bark waste of *Acacia crassicarpa* plantation wood in Indonesia. *J Wood Sci*. 63(4): 350-359. DOI 10.1007/s10086- 017-1633-4
[11] Ismayati M, Kamaluddin N. N, Nakagawa-izumi, A, Ohi H 2016 Toxicity and Feeding Deterrent Effect of 2- Methylanthraquinone from Wood Extractives of Tectona grandis on the Subterranean Termite Coptotermes formosanus and Reticulitermes speratus. Insect 7(4):63. DOI:10.3390/insects7040063

[12] Ismayati, M 2017 Studies on Wood and Bark Extractives as Natural Preservatives towards Subterranean Termites Doctoral's thesis. University of Tsukuba, Tsukuba, Japan.

[13] Ohara S, Suzuki K, Ohira T 1994 Condensed tannins from Acacia mearnsii and their biological activities. Mokuzai Gakkaishi 40:1363-1374

[14] Tascioglu C, Yalcin M, Troya TD, Sivrikaya H 2002 Termiticidal properties of some wood and bark extract used as wood preservatives. Bioresources 7(3), pp.2960-2969.

[15] Ohmura W, Doi, S, Aoyama M, Ohara S 2000 Antifeedant activity of flavonoids and related compounds against the subterranean termite Coptotermes formosanus Shiraki. J Wood Sci 46: 149–153