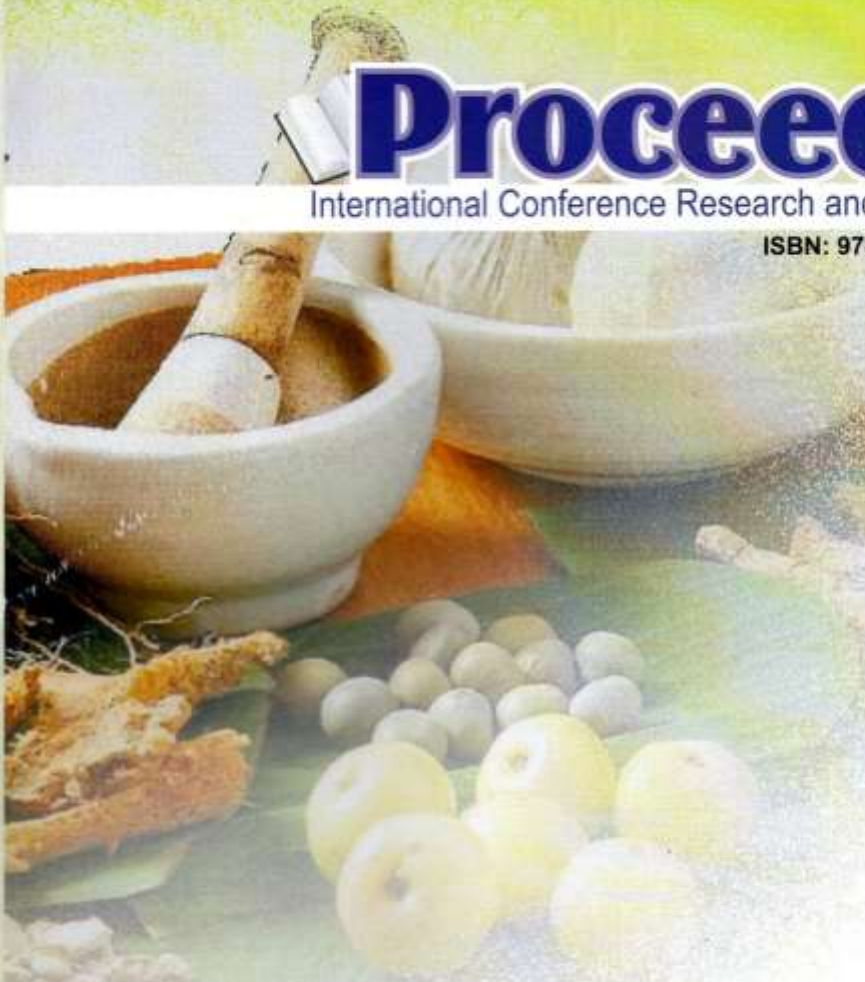


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PROCEEDING

INTERNATIONAL CONFERENCE
RESEARCH AND APPLICATION ON TRADITIONAL, COMPLEMENTARY,
AND ALTERNATIVE MEDICINE IN HEALTH CARE

Developing Traditional and Herbal Medicine
from customarily used to Scientifically Utilized

Surakarta, June 22nd-23rd, 2012
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Histopathological Picture of Liver of White Rat (*Rattus norvegicus*) that were Fed *Aloe vera* Gel after Exposed to Mainstream Kretek Cigarette Smoke

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Abstract

Cigarette smoke contains thousands of toxic components such as tar, nicotine, carcinogenic substances and free radicals that can be detrimental to health. High content of cigarette smoke free radicals, nitrosamines, cadmium and vinyl chloride causes increased risk of hepatotoxic effects and tissue damage. Aloe vera contains active substances, minerals and various vitamins that are nutritious, and act as hepatic protector. This study aimed to determine the effect of Aloe vera gel administration on liver histopathological picture of *Rattus norvegicus* that are exposed to mainstream kretek cigarette smoke. The design of research was pure experimental design by provision of treatment for 32 days. Research subjects were 24 male rats (*Rattus norvegicus*) Sprague Dawley strain, three months old, weight 200-300 g, divided into four groups: control group, exposed to mainstream kretek cigarette smoke (2 x 15 puff/day) group, the provision of Aloe vera gel (2x1.5 ml/200grWB/days) group, exposed to mainstream kretek cigarette smoke and provision Aloe vera gel group. One Way Anova statistical test followed by Duncan's Test against score of liver cell damaged show significant differences between the groups. It was concluded that Aloe vera gel treatment has a positive effect on liver histopathological picture of *Rattus norvegicus* which are exposed to mainstream kretek cigarette smoke.

Keywords: mainstream cigarette smoke, Aloe vera gel, liver histopathologic

INTRODUCTION

Smoking is a bad habit that has been carried out by people. Although the negative impact of smoking on health is known but in fact the smoking habit continues to this day. It is known that cigarette smoke contains tar, nicotine and many more substances or other substances that are harmful to health, such as CO, NO, NO₂, and free radicals (Eiserich et al, 1993). Cigarettes also contains nitrosamines, cadmium and vinyl chloride which causes increased risk of hepatotoxicity effects (Asfan, 2002).

Based on raw materials and its packaging, we can distinguish different types of cigarette, including (white) cigarette and kretek cigarettes. The one special type of cigarettes that are manufactured in Indonesia is kretek cigarette (Sitepoe, 2000). Kretek cigarette is most widely consumed in Indonesia (64.5%) (Risksesda, 2007).

Indonesia is rich in plants that are beneficial to health. *Aloe vera* is one of them. *Aloe vera* contains a few minerals such as Zn, K, Fe, Ca, Mn, Cr, Se, and vitamins A, B1, B2, B12, C, D and E, inositol, folic acid, kholin and 17 kinds of

amino essential amino acids (Anonym, cit Astuti, IIP, 2004 & Fertiasari, R, 2011)

MATERIAL AND METHODS

The main materials used in this study was the gel of *Aloe vera* and the filter kretek cigarette from 1 brand. This study was a pure experimental study using post only controlled group design. The subjects of this study were 24 white rats (*Rattus norvegicus*) male, Wistar strain, divided into 4 groups: control group, the group exposed mainstream smoke of cigarettes, the group given *Aloe vera* gel, and the group exposed mainstream smoke of cigarettes and given *Aloe vera* gel.

The *Aloe vera* gel dose given was 1.5 mL/200 g BW/treatment. *Aloe vera* gel obtained from the inside of the leaf was transparent, after being separated from the latex. Mainstream cigarette smoke cigarettes given by installing pipe on the filter, followed by sucking and blowing smoke with chip blower, and then put it into the mouth of rat. This was repeated 15 times on each treatment to each rat. *Aloe vera* gel treatment provision and mainstream smoke cigarettes given 2 times daily for 32 consecutive days.

On day 33 after treatment, the rats were killed and liver organ was taken. Liver histology preparation of organs was made by the method of paraffin blocks with hematoxylin eosin staining technique. Further, changes were observed on liver cells (hepatocytes). Microscopic observations to the preparations were done with 40x10 magnification, on 5 fields of vision, each field of view 40 hepatocytes observed. The changes of hepatocytes histologic features (parenchymatous degeneration, hydropic degeneration and necrosis) were observed and given scores.

Data were analyzed with One Way ANOVA followed by Duncan Multiple Range Test. For additional data, the liver weight of rat was measured

RESULTS AND DISCUSSIONS

The results showed that the liver in the **group** of cigarettes underwent the most **severe** degree of tissue damage (score **highest** liver damage) and much different from the histological features of the control **group** (figure 1, 2 and table 1). These results were consistent with the results of Abidin (2002) which examined the influence of White Cigarette smoke and Kretek Cigarettes exposure on histologic liver. In these studies the results showed that histologic features of liver of rats exposed to cigarette smoke showed a significant difference when compared with the control group. Kretek cigarette smoke contains high levels of free radicals. Cigarette smoke can disrupt the balance between antioxidants and free radicals in the body resulting in oxidative stress that can cause damage to body cells, including liver cells. Free radicals can damage macromolecules such as damaging the cell membrane lipids, DNA, proteins that cause cells to oxidative stress (Valko, et al., 2006).

The body is exposed to free radicals, both from the metabolic processes in the body, as well as free radicals from the outside. Exposure to cigarette smoke can cause an imbalance between antioxidants and free radicals. In this state the body needs antioxidants from outside to prevent continued damage by free radicals tissue. One of the many plants that contain antioxidants is the *Aloe vera*. Morsy (1991) states that in the *Aloe vera* gel contains nutrients: carbohydrates, fat, protein, vitamin A, vitamin C, thiamine, riboflavin, niacin, calcium and Fe.

In this study, *Aloe vera* gel gave positive effect on histopathologic features of the rats' hepatocytes. It can be seen in the rats exposed to mainstream cigarettes smoke and the ones given *Aloe vera* gel the lower damage scores compared to the group that presented mainstream cigarette smoke (without *Aloe vera* gel). This was due to *Aloe vera* gel contains a number of antioxidants such as vitamins A, B, E, C and minerals.

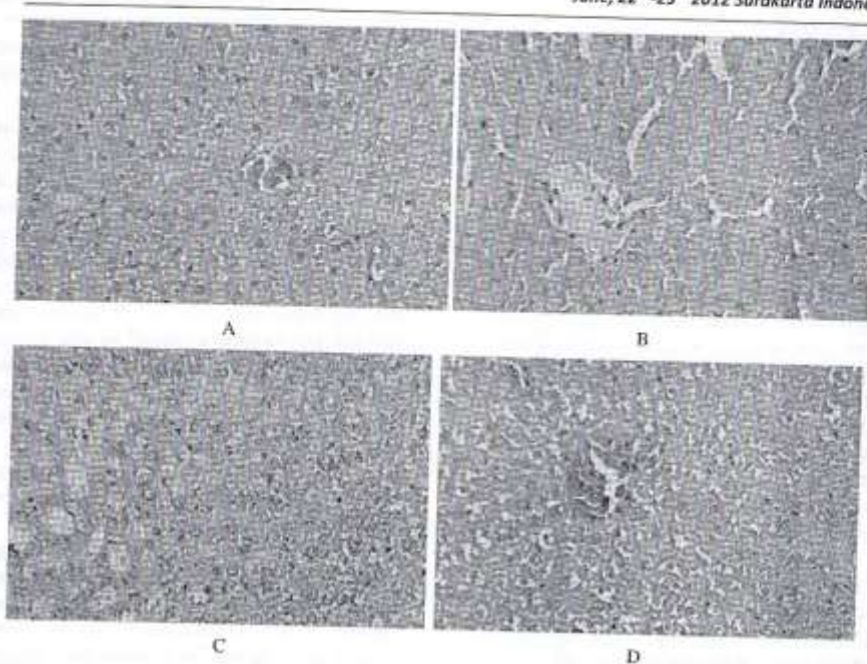


Figure 1: Overview of liver histology (HE; 40x10): (A) control group, (B) groups of *Aloe vera*, (C), *Aloe vera* group and cigarettes., (D) groups of cigarettes

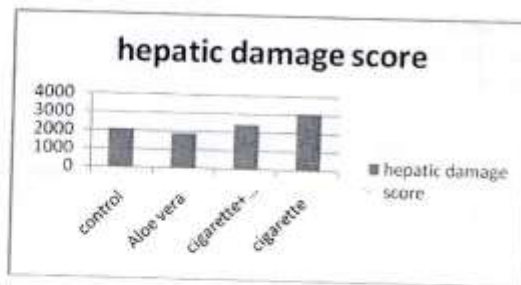


Figure 2: Hepatic damage score

Table 1: Hepatic Damage Score ± SD

Number	Treatment group	Average ± SD
1	Control	2105.00 ± 132.29 ^a
2	<i>Aloe vera</i>	1907.50 ± 96.89 ^a
3	Cigarette + <i>Aloe vera</i>	2455.83 ± 141.86 ^b
4	Cigarette	3095.00 ± 256.18 ^c

Note: the above numbers are the results of the ANOVA statistical test sig.0.00. a, b, c.: indicates a significant difference in post hoc Duncan test at 95% confidence level

Niacin which has protective properties for the body is able to prevent the formation of cell membrane damage. Niacin is a coenzyme, which has role in tissue integrity. Vitamin A is the main instrument in improving the integrity and permeability of cell membranes. Vitamin C has the ability as a free radical scavenger to suppress the free radicals that will attack the lipids. It can directly react with superoxide and hydroxyl anion. Vitamin C can also act as secondary antioxidant by maintaining a reduced glutathion that is very important for the body to ward off free radicals. Vitamin E, is a vitamin that acts as an antioxidant for the body, preventing damage and enhancing repair of cell membranes by non-enzymatic pathway (Harjasmita, 2004).

In the group given only mainstream cigarette smoke, free radicals and toxic compounds of cigarette smoke that enters the body were only resisted by antioxidants of the body. If the antioxidants of the body are unable to resist it, it will cause tissue damage. This is reflected in the description

of tissue damage in the liver damage score of cigarette group that have the highest score among all experimental groups.

The positive impact of the provision of *Aloe vera* gel on liver tissue was also seen in the group of *Aloe vera* which has a lower damage scores than the control group, although statistically the difference was not significant. *Aloe vera* has a natural content of hepatoprotective activity, making it possible to reduce tissue damage caused by oxidative stress in the body. This is consistent with findings by Woro et al (2002) which states that the water extract of *Aloe vera* has proven hepatoprotective activity.

The results of measurements of liver weight to groups of this study can be seen in figure 3 and table 2.

The results of rat liver weight showed no significant differences between groups. The difference observed in these figures is the normal variability of liver weight and can not be inferred as a result of the effect of giving the treatment.

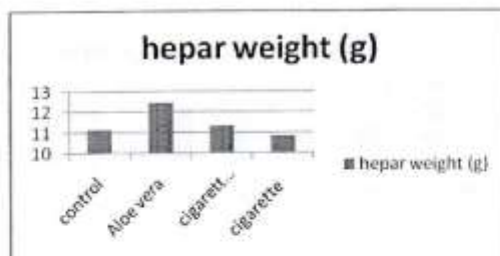


Figure 3. Hepar weight

Table 2. Average of Hepar weight \pm SD

Number	Treatment group	Average \pm SD
1	Control	11.18 \pm 0.56
2	<i>Aloe vera</i>	12.46 \pm 0.55
3	Cigarette + <i>Aloe vera</i>	11.36 \pm 0.56
4	Cigarette	10.83 \pm 0.81

Note: The Anova statistical test result shows there were no significant differences between groups ($p=0.994$)

CONCLUSION

It was concluded that *Aloe vera* gel treatment has a positive effect on liver histopathological picture of white rat (*Rattus norvegicus*) which are exposed to mainstream kretek cigarette smoke.

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