

# Effect of Robusta Coffee to Multiple Organ Histopathological Change on Rats Strain Wistar

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## Abstract

Coffee beans as beverages that contains acrylamide that formed in roasted process. Acrylamide can make multiple organ dysfunction on rats. The study aims to prove the effect of Robusta on Morphological changes of Hepatocytes, colonic mucosa, and prostatic acinar on Male rats . True experimental post test only control group design. 24 samples of male rats clustered in 5 groups; control group and treatment group were given steeping doses of coffee (0.36ml, 0.72ml, 1.44ml, and 2.16ml). Analysis of the data using *One-Way ANOVA* showed that there are significant value 0.000. Steeping of robusta coffee (*Coffea canephora var. Robusta*) could cause morphological changes of hepatocytes cell, colonic mucosa, and prostatic acinar in male rats (*Rattus novergicus Strain Wistar*).

**Keywords:** Robusta coffee, Morphological change.

## 1. Introduction

Coffee can be classed as a beverage psikostimulant that will cause people to stay awake, reduces fatigue, and makes happier. Therefore, it is not surprising that coffee became a favorite drink in the world, especially for men (Favrod-coune and Broers, 2010). Coffee beans contained chlorogenic acid and caffeine which is one of the main antioxidant (Alexander Y, Yakov Y, Jing Y W, et al.2013).

The coffee roasting process to go through before they can be consumed. In the roasting process will form acrylamide compound. Acrylamide is genotoxic and carcinogenic in animals. International Agency for Research on Cancer (IARC), the US Environmental Protection Agency (EPA), the Food and Drug Administration (FDA), as well as The National Toxicology Program has classified acrylamide as a substance that may cause cancer or potentially carcinogenic to humans (Group 2A) (Yahdiana, 2006).

Results of a meta-analysis study revealed that coffee consumption may reduce the risk of some cancers such as bladder cancer, breast cancer, pharyngeal cancer, colorectal cancer, endometrial cancer, liver cancer, and prostate cancer (Yu et al., 2011). On the other hand, several studies have shown that the content of acrylamide in coffee which has undergone a roasting process can cause some cancers (Belgrade, 2012).

Therefore, researchers wanted to see the effect of coffee on some of the strain Wistar rat organs, namely the colon, liver, and prostate.

## 2. Material and Method

### 2.1. Green Supply Chain Management

This research is true experimental using Post Test Only Control Group Design. To assess changes in memory function in male Wistar strain rats using the Morris Water Maze, an to assess histopathological changes in the brain in male Wistar rats.

This study using rats (*Rattus novergicus Wistar strain*) healthy male with a lifespan of 10-12 weeks and weigh 180-250 grams counted 25 animals were divided into five groups and each group a total of 5 rats. Group 1 (negative control) are a group of rats fed BR-1 and was not given any treatment. Group 2, 3, 4 and 5 are a group of rats given steeping robusta coffee (*Coffea canephora var. Robusta*) with each dose (0.36 ml, 0.72 ml, 1.44 ml, 2.16 ml) do for 28 days. Prior to the treatment, the mice conducted the process of adaptation. After treatment the mice off to assess histopathological changes in the rat colon, hepar and prostat illustration. Histopathologic changes were assessed is the presence of atypical cell on mucosa colon, hepatocyte necrosis, and prostatic acinar proliferation. Observations were made in 5 visual field with 400x magnification.

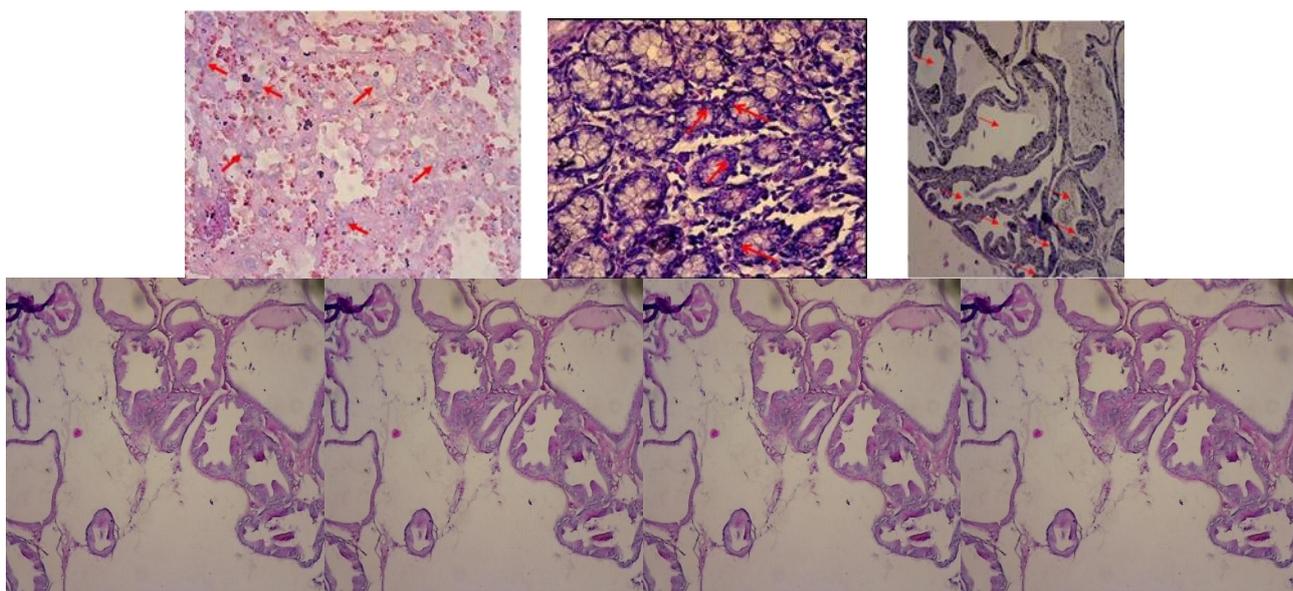
### 3. Result

**Table 1.** Number of atypical cell on mucosa colon, hepatocyte necrosis, and prostat acinar proliferation on each group

Group	Number of atypical cell on mucosa colon	Number of hepatocyte necrosis	Number of prostatic acinar proliferation
control	2,15	62,4	0
T1 (0,36 ml)	11,6	55,65	3
T2 (0,72 ml)	20,8	29,15	4
T3 (1,44 ml)	55,10	14,75	5
T4 (2,16 ml)	65,10	8,45	7
P (Annova)	0,000	0,000	0,000

Note : T : treatment group; p is significancy of Annova test (significant if  $p < 0,005$ )

Based on table 1 and picture 1, there are significancy of effect robusta coffee to histopathological change on mucosa colon, hepatocyte cell, and acinar prostat. Atypikal the colon mucosa cells characterized by enlarged cell nuclei, mild pleomorphic, with scant cytoplasm. While necrosis of hepatocytes characterized by nuclei kariolysis of hepatocytes cell. Prostatic acinar cell proliferation characterized by the addition of number acinar cells, which is described by the tufting on acinar.



**Picture 1:** Histopathological change on hepatocyte (A), mucosa colon (B), prostatic acinar proliferation (C). (HE, mag 400x)

### 3. Conclusion and Conclusion

This study proves that there are differences in the number of cells undergoing proliferation in rats male Wistar strain between the negative control group to the treatment group. In the group of male Wistar strain rats by administering doses of

This study proves that there are differences in the number of cells undergoing proliferation in rats male Wistar strain between the negative control group to the treatment group. In the group of male Wistar strain rats by administering doses of robusta coffee steeping at 2:16 ml / 200 kg has more results prostatic acinar cell proliferation compared to the other dose treatment group.

Coffee has the effect of increasing the hormone androgen. Androgen hormones have an important role in the development of the prostate gland. Growth factor effect on epithelial-stromal interactions in the physiology of the prostate gland. Androgens and Growth Factor influence the proliferation, differentiation and function of cells - the cells of the prostate gland. Some of the epithelial growth factor mediates interaction stroma of the prostate gland. TGF  $\alpha$ , EGF, basic fibroblast growth factor, keratinocyte growth factor and insulin growth factor stimulates the growth of cells - epithelial cells of the prostate gland. TGF $\alpha$  expression can be found in the ventral prostate gland normal mice. TGF $\alpha$  also play a role in prostate cancer. Expression of TGF $\alpha$  found in prostate cancer. but not reported in prostate cancer cell line LN CaP and PC-3, TGF $\alpha$  expression is directly regulated by androgens. Regulation and expression of growth factors is complex and does not include only by androgen, but also by the cooperation of the production factors - factors locally in the prostate gland. The theory according to this study that the greater the dose of coffee is provided, the more prostate acinar cell proliferation.

The study also proved that there is a difference in the number of normal cells in the liver hepatocytes white male rats between the negative control group to the treatment group. In the group of white male rats with dosing of robusta coffee 2,16ml / 200g BB rats have on the calculation normal hepatocyte cell number in the liver at least compared to the other dose treatment group that obtained the results of the calculation of average number of normal hepatocyte cells of 8.45. This is consistent with previous research by Bhara in 2009 stating that the coffee had an effect on hepatic histology white male rats which indicates a change in the cell in the form of edema, hydrophic degeneration, and degeneration parenchyme.

The mechanism of toxic substances such as acrylamide that is carcinogenic would cause injury through oxidation-reduction reaction that will increase the Reactive Oxygen Species (ROS), cells are exposed to injury can not eliminate the water that accumulate in the cell so that the cell swelling, constant exposure cause injury irreversible. Injured cells could then be experiencing tear the plasma membrane and the cell nucleus changes that cause cells to die. Necrosis is a pathological process irreversible after cell injury caused active inflammatory mediators such as TNF, IL-1

that activate stellate cells into collagen-producing cells myofibroblast excessive, this triggers the onset of fibrotic tissue. The next stage may occur fibrogenesis in chronic exposure can lead to cirrhosis of the liver which is the main risk factor for the occurrence of liver cell carcinomas (Robbins, 2014).

In addition, this study also proves that there are differences in the number of atypical cells in the epithelium of the colon in male rats between negative control group with treatment group. This is consistent with previous studies by Tanseri L 2010, which states that acrylamide in coffee shows activity as a trigger tumors in rats and mice which reactive conduction pathways of acrylamide can disrupt the intracellular signal that induces changes in cell function.

Steeping coffee robusta contained acrylamide which is the end product of the process of roasting coffee, in the form of a crystal clear, odorless, easily soluble in water and in many other organic solvents, but insoluble in benzene and heptane (Harahap Y, 2006; Tanseri L, 2010) , a cup of acrylamide concentration is from 0.32 to 1.46 g / 30ml in 6 g of coffee. Acrylamide is a substance that is harmful and potentially cause cancer in about 2% of cases each year in the world. Acrylamide is usually found in foods that are processed using high temperatures above 150°C (Herman S and Adawiyah R, 2010). Acrylamide is not formed at a temperature below 120oC (Harahap Y, 2006).

Acrylamide is carcinogenic mechanism that will lead to changes in the chemical structure of DNA, causing DNA damage in the colon mucosa (Tanseri L, 2010). DNA damage activates p53 to help repair DNA by stopping G1 and induce gene DNA repair through increased gene transcription of inhibitors of cyclin-dependent kinase CDKNIA (p21) and gene GADD45, DNA repair that successfully allows the cell to continue the cell cycle if DNA repair fails, p53 will trigger apoptosis or senescence. In cell loss or a mutation of the TP53, it will not induce DNA damage cessation of cell cycle or DNA repair and cell with genetic damage will still proliferate, to then grow into malignant neoplasms (Robins, 2014).

In the colonic mucosa tumor suppressor gene mutations occurred in one allele of the APC 5q21. The second allele mutation of the APC gene was found in colonic mucosa at risk, mucosal epithelium will be found in the form of atypical cells. The cause of these atypical cells could be due to a mutation, infection or aging. Furthermore, the K-RAS gene mutation occurred in the adenoma 12p12, p53 gene mutations occur in developing adenomas that would later become carcinoma. If the p53 gene is not mutated then adenoma still be adenomas aging as induced by oncogenes assisted by wild-type p53 (Robins, 2014).

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