

Radioprotective of Roselle Tea in Recurrent Radiodiagnostic Ionizing Radiation on Peripheral Hemogram Characteristics

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Abstract

The aim of this study was to describe peripheral hemogram characteristics of mice treated with Roselle tea prior to exposure of recurrent radiodiagnostic ionizing radiation. Eighteen adult ddy male mice of Mus musculus were divided into three groups, i.e. control group (K, n=6), ionizing radiation group (P, n=6), and tea of roselle radiation group (RT, n=6). Blood samples were collected from blood venous at sinus retroorbitalis of anesthetized mice. These blood samples were examined for the numbers of leucocyte (WBC), erythrocyte (RBC), hemoglobin (Hb), hematocrit (PCV), RBC volume (MCV), Hb weight (MCH), and Hb concentration (MCHC). Blood samples were collected from mice which had been exposed of ionizing radiation for 2 weeks and 4 weeks periods. The result showed that there were no significant differences ($P>0.05$) on WBC, MCH, and MCHC. However, we found differences ($P<0.05$) on hematology characteristics for RBC, Hb, PCV, and MCV of mice after 2 weeks exposure of ionizing radiation.

Keywords: *hematology, ionizing radiation, roselle tea.*

INTRODUCTION

X-rays were first discovered on November 8, 1895 by the German physicist Wilhelm Conrad Roentgen. Soon afterward, x rays was used as a diagnostic tool. During its first 100 years of discovery, x-rays were widely used for the benefit of the medical picture of the disease (McCurnin & Bassert 2006). Nowadays, X-rays is commonly used to diagnose or as a component of therapy for a wide range of disease conditions. Radiodiagnostic were performed by imaging to diagnose degree of the disease such as the primary tumor, and subsequent metastasis can be visualized non-invasively by whole

body imaging through the patient (Bouvet *et al.* 2002; Jeong *et al.* 2003). However, this procedures can develop to residual (or long-term) normal organ or tissue injury (Mauch *et al.* 1995). Ionizing radiation of X-ray is a hazard ionizing radiation. It causes damages on the exposed tissues (Thrall 2002). Low dose of ionizing radiation exposure from X-ray causes differences or destroys tissue (McCurnin & Bassert 2006).

Currently, an alternative mechanism to reduce toxicity to normal tissues is the use of radiation modifiers/protectors, agents that when present prior to or shortly after radiation exposure alter the response

of normal tissues to irradiation (Citrin *et al.* 2010). Many naturally occurring compounds found in medicinal plants, herbs, and spices have been shown to possess antioxidant activities against ionizing radiation. *Hibiscus sabdariffa* L. (Roselle), a medicinal herb commonly uses to make drink and pickle, is used in folk medicine in the treatment of hypertension, liver diseases, and fever (Wang *et al.* 2000; Odigie *et al.* 2003; Akindahunsi & Olaleye 2003). This study aimed at describing peripheral hemogram characteristics of mice by providing natural Roselle tea prior to exposure of recurrent radiodiagnostic ionizing radiation.

MATERIALS AND METHODS

Eighteen male ddy mice 6-8 weeks age with body weight 30-40 gram were used as experimental animals. Mice were caged by plastic box covered by wire (30x40 cm) with commercial foods and *ad libitum* water. The mice were acclimatized for two weeks prior to ionization radiation. They were divided into 3 groups of treatment, i.e. control non-ionizing radiation group (K, n=6) 0.9% NaCl 0.2 ml orally, ionizing radiation group (P, n=6) 0.9% NaCl 0.2 ml orally and Roselle tea with ionizing radiation group (RT, n=6)

500gram roselle/100ml aquadest 0.2 ml orally.

Roselle tea was prepared by boiled system at 90°C of temperature for 15 minutes, then cooled to 30°C (Fig 1B1-2). The ionizing radiation exposure were performed by radiodiagnostic portable X-ray (VR-1020, MA Medical Corp. Japan) as tool model. Total dose of ionizing radiation for mice were 1.6 mSv/2 weeks and 3.4 mSv/4 weeks, performed every 2 days after being given saline or tea orally by 0.2 mSv/exposure radiation dose (Fig 1B3). Total Body Radiation (TBR) radiography exposure setting for focal focus distance (FFD) by 100 cm, kilovoltpeak (kVp) by 80, and mili Amphere second (mAs) by 20 produced 0.2 mSv of ionizing radiation on each exposure (Noviana *et al.* 2010). The ionizing radiation exposures were measured by digital dosimeter MYDOSE miniTM (ALOKA CO., LTD Tokyo Japan).

Blood samples were collected from blood venous at sinus retro-orbitalis of anesthetized mice (Thrall 2004). These blood samples were examined for the numbers of erythrocyte (RBC), leucocyte (WBC), hemoglobin (Hb), hematocrit (PCV), RBC size (MCV), and Hb concentration (MCHC). Blood samples were col-

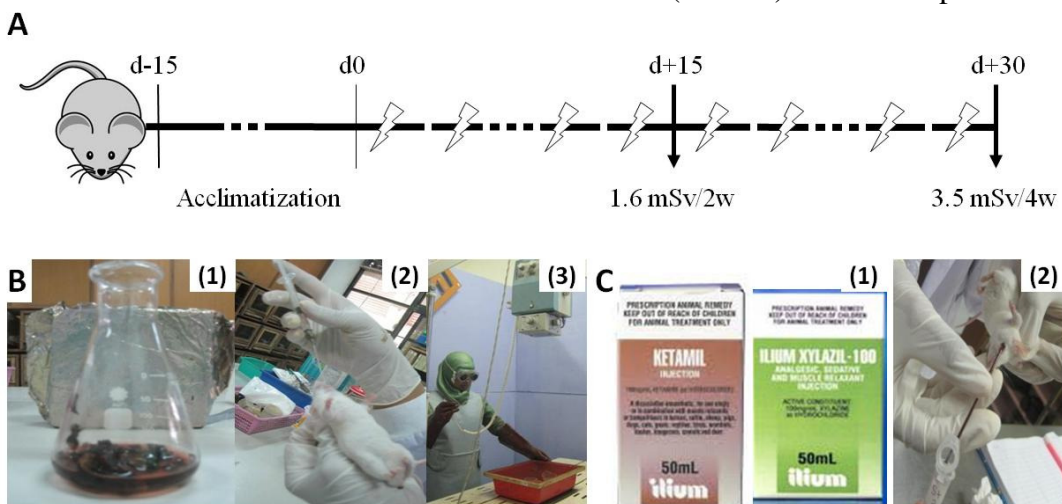


Figure 1. Design of study, total body radiation (TBR) by radiography, and blood collection. A. Design of study; B. Total body radiation (TBR) procedure, (1) Roselle tea, (2) gavaging prior to radiation, (3) TBR radiography; C. Blood collection procedure, (1) Ketamine and Xylazine anaesthetic, (2) blood collection. d = days; mSv = mili Sievert; w = weeks.

lected from mice which had been exposed of ionizing radiation for 2 weeks and 4 weeks period (Fig 1C1-2).

The obtained data were expressed as mean \pm standard deviation of means ($\bar{x} \pm SD$). A One-Way Analysis of Variance (ANOVA) was used to compare the means of the studied groups with post hoc Duncan test at 5% for those results where a significant difference was indicated. The

Statistical Package for Social Sciences (SPSS[®]) version 13 for Microsoft[®] Windows[®] statistical software was used.

RESULTS AND DISCUSSION

The RBC count of Roselle tea was higher than radiation and control group after receiving 1.6 mSv total body radiation (TBR). In other hand, no differ-

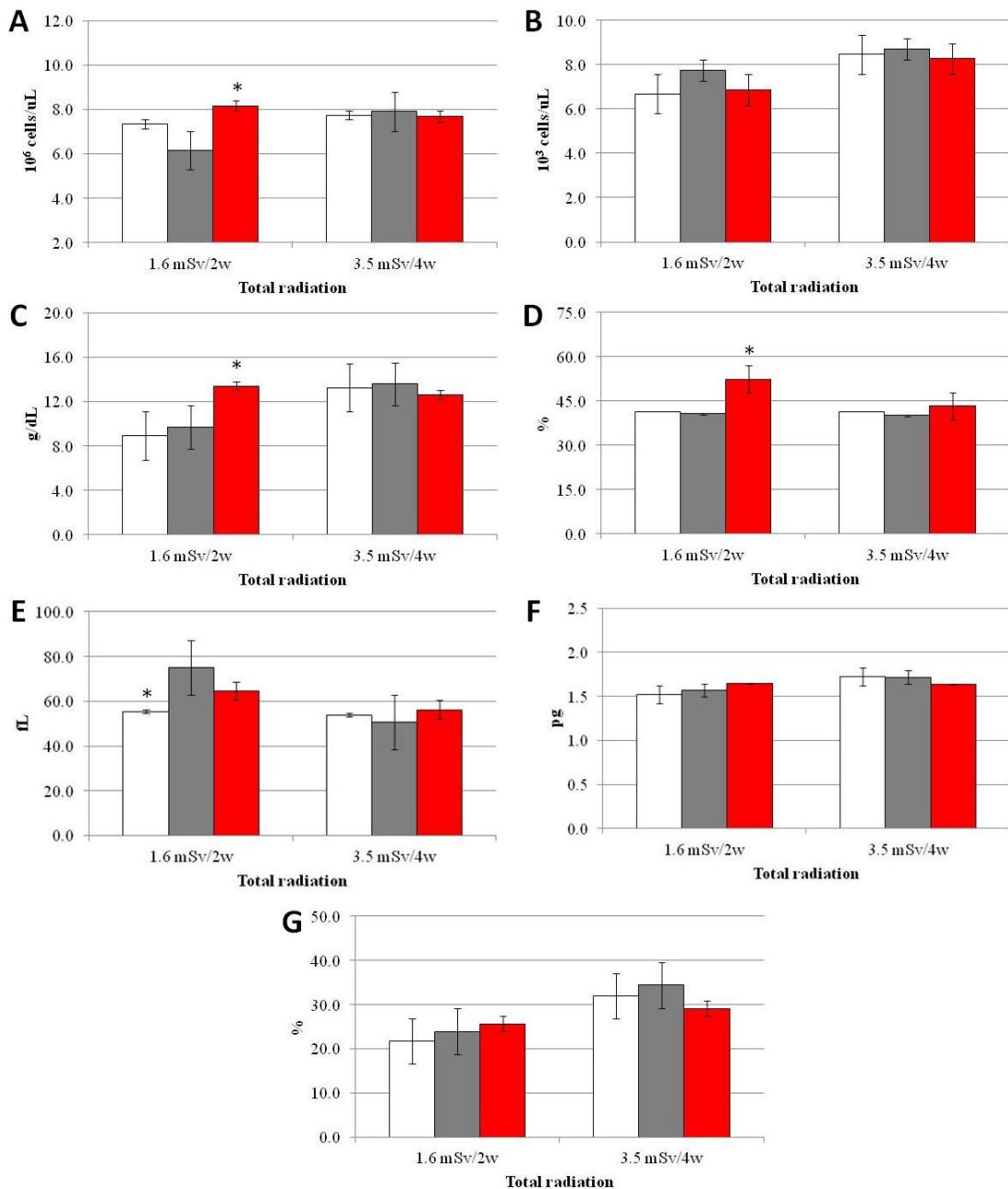


Fig 2. Design of study and whole blood characteristic. A. Red blood cell (RBC); B. Leucocyte (WBC); C. Hemoglobin (Hb); D. Hematocrite (PCV); E. RBC volume (MCV); F. Hb weight (MCH); G. Hb concentration (MCHC); ⚡ = radiation exposure from radiodiagnostic radiography; □ = control; ■ = radiation; ■ = roselle tea; * = $P < 0.05$; mSv=mili Sievert dose of radiation; RBC, WBC, Hb and PCV data from Ningsih *et al.* (2011).

ences between groups after receiving 3.5 mSv TBR (Fig 2A). The WBC count, Hb weight, and Hb concentration (MCHC) showed that no difference in all group and TBR (Fig 2B,F,G). The Hb and PCV data showed that only on Roselle tea group after receiving 1.6 mSv of TBR higher than other group (Fig 2C,D). However, RBC size on both of radiation and Roselle group were higher than control (Fig 2E).

Parameter measurement of total peripheral blood at 1.6 mSv of radiation for 2 weeks showed that there were disturbances in the blood parameters. While the total radiation was 3.5 mSv after 4 weeks of the condition in which the body had radio-adaptation response. The RBC values decreased in the radiation group after receiving 1.6 mSv total radiation exposure, indicating the signs of anemia. Anemia occurs due to the reduced number of RBC from the normal threshold values caused by exposure of ionizing radiation. The MCV increased in radiation group than control after receiving 1.6 mSv total radiation exposure. This condition was caused by low levels of vitamin B12 and folic acid in the peripheral blood. This condition occurs probably due to impaired absorption of nutrients in the digestive system and lead to macrocytic anemia. Supplementation of Roselle tea can repair this condition close to normal as the control group. Because MCH value in normal range conditions, so it is called as normochromic macrocytic anemia (NMA). The NMA is a regenerative anemia and become normal if the causative factor was eliminated (Thrall 2004).

The result showed that ionizing radiation energy has negative effect for hematopoietic process (Wambi *et al.* 2009). Occupational exposures of ionizing radiation from medical diagnostic imaging give 35 % contribution of about 0.8 mSv (DMP 2011). The long-term effects of low-dose exposures from repeated radiodiagnostic imaging may be real and should be given serious consideration (Tucker 2008).

Ionizing irradiation clearly induces multiple changes both within cells and in their microenvironment (Little 2000; Barcellos-Hoff *et al.* 2005). Irradiation also causes a high degree of in vivo hydroxy radical generation by hemolytic cleavage of body water or from the endogenous hydrogen peroxide formed by reduction of the superoxide anion. The hydroxyl radical is the major cytotoxic radical than others. The interaction of these radicals with phospholipid structures induces the peroxidation processes that increase hydroxyl radical activity in the DNA oxidative damage. Under these oxidative stress conditions, although radiation therapy is needed to patient with cancers, exogenous agents with a radical scavenging capacity need to be used to minimize the normal cell damage (Castillo *et al.* 2000)

The supplementation with antioxidant vitamins and minerals may have a protective effect against X-ray-induced damage. Flavonoids compound shows 20 times more powerful antioxidant activity than vitamin in the lipoprotein oxidation model (Craig 1999). Roselle contains vitamin C more than threefold of black grape, nine fold of citrus lime, and tenfold of star fruit (Widyanto & Nelistya 2008). Phytochemical screening and structural analysis has identified a class of flavonoids called anthocyanin as responsible for antioxidant activities (Passamonti *et al.* 2003; Hou *et al.* 2005). Supplementation of Roselle tea has been able to maintain conditions of peripheral blood parameters in the threshold of range normal condition as control group. The ability of Roselle tea to protect blood cells against damage caused by ionizing radiation called as radio-protective effect.

CONCLUSION

Based on the result we conclude that Roselle tea have potency of radio-protective effect from exposure of ionizing radiation of radiodiagnostic radiography procedure in mice (*Mus musculus*).

ACKNOWLEDGMENT

This study was supported by Directorate of General Higher Education (DGHE) of Indonesia in Student Creativity Program 2010 collaborated with Competitive Grant also from DGHE (28/I3.24.4/SPK/PD/2010) in the same year.

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