**Amaranthus tricolor Linn** PRESERVES THE THICKNESS OF SEMINIFEROUS TUBULES IN MICE (Mus musculus) INDUCED BY LEAD ACETATE

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**ABSTRACT**

Spermatogenesis is a cyclic and continuous process which starts at puberty and carries in throughout the majority of male’s life span. The purpose of this research is to determine the protective effect of red spinach (Amaranthus tricolor Linn) on male reproductive system, which would preserve the thickness of seminiferous tubules in mice induced by lead acetate. Thirty mice were devided into 3 groups, K0 as the control group was given placebo, K1 as the treatment group was given lead acetate 75 mg/kgBW/day and placebo, K2 as the treatment group was given lead acetate 75 mg/kgBW/day and 382 g/kgBW red spinach. The conclusion of this research is that red spinach amaranth can preserves the thickness of seminiferous tubules in mice induced by lead acetate. (FMI 2013;49:226-228)

**Keywords:** Amaranthus tricolor Linn, seminiferous tubules, lead acetate

**INTRODUCTION**

Spermatogenesis is a cyclic and continuous process which starts at puberty and carries in throughout the majority of male’s life span (Dadoune 2007). This process takes place in the seminiferous tubules which is a layer consisting mixture of Sertoli cells and spermatogonia. The availability of testosterone produced by the Leydig cells becomes the main prerequisite. Spermatogenesis that goes well will be a determining factor for the man fertility. Lead (Pb) toxicity has been known for a long time and mostly become the research object in industrial toxicology. One cause of men infertility with unknown etiology was due to lead exposure (Vigeh et al 2011). Humans can be exposed with lead from environment and occupation. The effect of lead on the reproductive system is complex and involves diverse mechanisms. Lead exposure will eventually lead to impair fertility by decreasing the amount of spermatozoa, the motility of spermatozoa, and the pathological changes in the histology of the male reproductive system.

Indonesia has many medicinal plants which are beneficial to health. One of them is red spinach/amaranth (Amaranthus tricolor Linn), known as a medicinal plant in Amaranthaceae family. This plant grows all over the year and can grow in a hot climate. Red amaranth has been widely used as blood cleaning medicine, powerful medicine, dental pain medicine and cough medicine. Bioactive contents in the red amaranth are betalain, carotenoids, vitamin C, flavonoids, and polyphenols. The purpose of this research is to determine the protective effect of red spinach on male reproducte system, which preserves the thickness of seminiferous tubules in mice induced by lead acetate.

**MATERIALS AND METHODS**

The research subjects were mice (Mus musculus) aged 3 months old and weighed 29-31 grams. Sample size was 30 mice. This research used experimental design, Posttest Only Control Group Design. Mice qualified
were divided into 3 groups and treated differently. The first group was the control group who received distilled aqua and placebo (Na CMC 0.5 %). The second group was the treatment group given lead acetate (dose of 75 mg/kg weight per day) which was dissolved in distilled aqua and placebo in the form of CMC Na 0.5 %. The third group was the treatment group given lead acetate (dose of 0.075 g/kg weight per day) which was dissolved in distilled aqua and red amaranth juice (dose of 382 g/kg weight per day), also dissolved in CMC Na 0.5 %. Determination of lead exposure doses based on previous research by Widarti (2006), whereas the dose of red amaranth juice was equivalent to the dose of Trolox (water soluble vitamin E) to get the antioxidant capacity of 3.7 mmol for adults weighing 294 grams (Cao et al 1998), which had been converted for mice. The whole group was given treatment for 4 weeks and at the end of the fourth week, mice were sacrificed, and testes were taken for histological examination in the form of thick measurement seminiferous tubules.

RESULTS

Table 1. Thickness measurement results of seminiferous tubule (nm)

<table>
<thead>
<tr>
<th>Variable</th>
<th>K0 (n=10)</th>
<th>K1 (n=10)</th>
<th>K2 (n=10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>The thickness of seminiferous tubule (nm)</td>
<td>58.76 ± 3.89</td>
<td>47.18 ± 4.1</td>
<td>60.23 ± 2.69</td>
</tr>
</tbody>
</table>

All data were showed in mean ± SD (Table 1). Data were analyzed using ANOVA followed by LSD. The results showed that the thickness of seminiferous tubule in K1 group decreased compared with K0 and increased in K2 (Table 1). This shows that the dose of lead given had damaged the male reproductive system of mice and red amaranth juice could repair it. Data were analyzed using ANOVA and showed significant results (p < 0.05). Analysis results using LSD showed that there were significant decreases between K0 and K1, K1 and K2 (p < 0.05) and there was no significant increase between K0 and K2 (p > 0.05).

DISCUSSIONS

The results showed that all of the dependent variables at K1 were lower than K0. It showed that the lead acetate treatment given had caused damage to the male reproductive system of mice. Sokol et al (2002) stated that lead was a toxicant for the male reproductive system. Seminiferous tubules were epithelium tissues composed of germ cells and Sertoli cells. Sertoli cells played very important role in maintaining germ cells and regulating a variety of important processes in spermatogenesis (Dadoune 2007). Tubular epithelial thickness indicated the number of spermatogenic cells that would eventually become mature spermatozoa. The decrease in thickness will affect quantitatively the decrease in the concentration of spermatozoa in the ejaculate fluid (Dadoune 2007).

The results were consistent with research conducted by Hamadouche et al (2009), in which the rats exposed to lead at a dose of 250 mg/l and 500 mg/l showed an increase in the tubular lumen diameter and a decrease in the number of spermatozoa from the epididymis, FSH and LH, and Sertoli cell number (Hamadouche at al 2009). Some researchers estimated that the mechanism of the toxicity of lead on reproduction was at the level of the hypothalamic-pituitary axis or a combination of direct damage to the gonads and hypothalamic-pituitary. Disruption of spermatogenesis occurred as a result of a decrease in the amount of serum testosterone, because rats being exposed by lead showed a decrease in the amount of serum testosterone, FSH and LH (Hamadouche et al 2009, Hsu et al 1998).

Lead exposure also caused oxidative stress which was indicated by the lipid peroxidation products (LPP) increasing in the testis, epididymis and pituitary (Hamadouche et al 2009). Peroxidative damage due to ROS had been postulated as one of the main causes of sperm function defective (Hsu et al 1998). Naha and Chowdhury Research (2005) concluded that lead can penetrate the blood testis barrier and cause accessories sex gland dysfunction, sperm morphology abnormality and oligospermia. Recent research suggested that in the blood testis barrier was found perotein transporter for some metals, including DMT1 and Znt1 which possibly played an important role in individual susceptibility to
Based on literature studies conducted, it was concluded that red amaranth contained flavonoids, betalain, vitamin C and vitamin A, which was a potent anti-oxidant. Herb flavonoids had many benefits, including increasing the amount of testosterone and dehydrotestosterone, preventing cell death and destructing mitochondrial membrane, also as an antimicrobial and antifungal (Esfandiari & Dehghani 2010). High diet in fiber, Fe, Ca, zinc, and phosphorus in red amaranth (Amaranthus tricolor Linn.) could be expected to decrease lead absorption in the gastrointestinal tract resulting in lower lead levels in the blood. The mechanism was probably through a competitive inhibitor of the Fe, Ca, zinc, and phosphorus with lead at different binding sites on the cell membrane or in intracellular (Center for Disease Control and Prevention 2007).

Red Spinach contained vitamin A, vitamin C, and flavonoids Betalain which were potent anti-oxidant. Hsu et al (1998) researched on mice exposed to lead acetate and then given vitamin C or vitamin E. The research showed sperm motility increasing and the capacity of sperm increasing to penetrate the ovum. The protective effect was not associated with lower levels of lead in the blood and sperm, but the ability of antioxidant effects of these materials to resist spermatotoxicity lead to the formation of spermatozoa due to ROS (Hsu et al 1998). Besides as antioxidants, Betalain and vitamin C were also expected to have the ability to form complexes with the metal so that the possibility of tissue and blood lead levels could also be reduced through that mechanism (Hsu et al 1998). Red amaranth contained folate which was known as a growth factor that stimulated DNA replication (Guyton & Hall 2006) and herb flavonoids that had many benefits, including increasing the amount of testosterone and dehydrotestosterone, preventing cell death and mitochondrial membrane destruction, and also as antimicrobial and antifungal (Esfandiari & Dehghani 2010). The things mentioned above might explain why the result of the dependent variable on K2 was higher than K0.

CONCLUSION

The protective effect of red amaranth on male reproductive system is preserving the thickness of seminiferous tubules in mice induced by lead acetate.

REFERENCES


Center for Disease Control and Prevention (2007). Toxicology Profile of Lead, Georgia, US Department of Health and Human Service


