The Potency of Hylocereus polyrhizus Peel Extract as Protector on Lead Acetate-Induced Testicular Toxicity in Mice

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Submission date: 21-Jun-2020 03:07PM (UTC+0800)
Submission ID: 1347291692
File name: otency_of_Hylocereus_polyrhizus_Peel_Extract_as_Protector_on.pdf (188.66K)
Word count: 1500
Character count: 8006

Indian Vet. J., September 2019, 96 (09) : 49 - 51

The Potency of *Hylocereus polyrhizus* Peel Extract as Protector on Lead Acetate-Induced Testicular Toxicity in Mice

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(Received : March, 2019 **72/19** Accepted : May, 2019)

Abstract

This study aimed to investigate the treatment of *Hylocereus polyrhizus* peel extract 250 mg, 500 mg, and 1000 mg/kg BW orally once in a day for 40 days to improve the thickness of seminiferous tubular epithelium, Leydig cells, and spermatogenic cells in mice which were exposed to lead acetate 30 mg/kg on the 5th day one hour after the the *Hylocereus polyrhizus* peel extract administration for 35 day. The *Hylocereus polyrhizus* peel extract administration significantly increased the thickness of seminiferous tubular ephitelium, number of leydig cells and spermatogenic cell in lead acetate-induced testicular toxicity. The *Hylocereus polyrhizus* peel extract could be used as the protector in lead acetate-induced testicular toxicity.

Key words: *Hylocereus polyrhizus* peel extract, Lead acetate, testicular cells, mice.

Lead a heavy metal pollutant in the environment which is toxic and causing reproductive disorders (Sudjarwo *et al*., 2017). This leads to increased oxidative stress by forming reactive oxygen species (ROS) such as superoxide ion (O_2^-), hydroxyl radical (OH^-) and nitric oxide (NO), and direct suppression of antioxidant reserves such as Superoxide Dismutase (SOD), Catalase and Glutathione Peroxidase (GPx) (Sudjarwo *et al*., 2019). *Hylocereus polyrhizus* is a unique fruit and the *in vitro* studies have revealed that *Hylocereus polyrhizus* extract has the potential to be an antioxidant (Tsai *et al*., 2019)

Materials and Methods

40 male mice divided into 5 groups: negative control (mice were given aquadest daily), positive control (mice were given lead acetate 30 mg/kg BW orally once a day for 35 days), and the treatment group (mice were given *Hylocereus polyrhizus* peel extract 250 mg, 500 mg, 1000 mg/kg BW orally once a day for 40 days, and on the 5th day, were given lead acetate 30 mg/kg BW one hour after *Hylocereus polyrhizus* peel extract administration for 40 days). On day 40, mice were sacrificed, and testicular tissues were fixed in formalin 10% to evaluate the thickness of the seminiferous tubular epithelium measuring the, number of Leydig cells and number of spermatogenic cells in histopathological study.

Results and Discussion

Table I showed in the lead acetate treatment group, the thickness of the seminiferous tubule epithelium and the number of Leydig cells of testis tissue were significantly decreased compared to the negative control. Treatment with *Hylocereus polyrhizus* peel extract at dose 1000 mg/Kg BW markedly increased the thickness of the seminiferous tubular epithelium and the number of Leydig cells which is significantly different from the positive control.

The thickness of the spermatogenic cells (spermatogonium, spermatid and spermatozoal cell) of testis were significantly decreased in positive controls compared to the negative control. Treatment with *Hylocereus polyrhizus* peel extract at dose 1000 mg/Kg BW markedly increased the spermatogenic cells (spermatogonium, spermatid and spermatozoa cell) in lead acetate treatment which was significantly in comparision to the positive control (Table II).

The histopathological changes in rat testis in the lead acetate treated group showed

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Table I: Effects of *Hylocereus polyrhizus* peel extract on thickness of seminiferous tubules and Leydig cells

Groups	Thickness of seminiferous tubular (μ/m)	The number of Leydig cells
Negative Control	141.5^{a} ± 16.5	36.3^{a} ± 4.9
Positive Control	113.5^{b} ± 12.1	23.2^{b} ± 5.6
H. polyrhizus 250 mg/kg BW	111.8^{b} ± 10.4	20.6^{b} ± 3.6
H. polyrhizus 500 mg/kg BW	121.9^{b} ± 18.9	24.1^{b} ± 4.5
H. polyrhizus 1000 mg/kg BW	136.7^{c} ± 14.7	30.5^{c} ± 4.2

^{a,b,c}Different superscript within each column differ significantly ($P < 0.05$)

Table II: Effects of *Hylocereus polyrhizus* peel extract on the number of spermatogenic cell

Groups	The number of spermatogonium cells	The number of spermatid cells	The number of spermatozoa
Negative Control	106.7^{a} ± 6.9	40.7^{a} ± 4.5	36.2^{a} ± 4.9
Positive Control	51.6^{b} ± 7.8	28.8^{b} ± 3.2	19.5^{b} ± 2.4
H. polyrhizus 250 mg/kg BW	60,1^{b} ± 5.9	30.6^{b} ± 2.7	20.5^{b} ± 5.5
H. polyrhizus 500 mg/kg BW	65.5^{b} ± 3.1	33.2^{b} ± 4.3	24.2^{b} ± 4.2
H. polyrhizus 1000 mg/kg BW	85.3^{c} ± 3.2	38.3^{c} ± 3.8	34.2^{a} ± 5.4

^{a,b,c}Different superscript within each column indicate significant difference between the means ($P < 0.05$)

the loss of spermatogenic cells and necrosis compared to the control group. The testicular damage (necrosis) was considered mild in the groups treated with *Hylocereus polyrhizus* peel extract (Fig 1).

Lead can induce oxidative stress via reactive oxygen species (ROS) generation, which has been reported as an important mechanism underlying lead induced-testicular toxicity (Gagan *et al*., 2012; Ali *et al*., 2018). Oxidative stress occurs when the generation of ROS increase and the scavenging capacity of antioxidants decrease in the cells. It has been reported that lead-induced over production of reactive oxygen species (ROS) or free radicals such as superoxide ion (O_2^-), hydroxyl radical (OH^-) and nitric oxide (NO), and consequently enhance lipid peroxidation, impairment of antioxidant enzymes activities, such as superoxide dismutase (SOD), Catalase (CAT), and glutathione peroxidase (GPx) (Sudjarwo *et al., loc cit*). In addition, free radicals are highly reactive to membrane lipids, protein, DNA and to be the major contributing factors to stress injuries and to cause rapid testicular cell damage (Xu *et al*., 2008; Sudajarwo *et al*., *loc cit*). It has been reported that extract have antioxidant activity and free radical scavenger. This suggests that *Hylocereus polyrhizus* peel extract inhibited the lead acetate induced-testicular toxicity, through its antioxidant activity.

Summary

The lead acetate administration significantly induces oxidative stress that an important mechanism underlying lead induced-testicular toxicity. While the *Hylocereus polyrhizus* peel extract has antioxidant activity which could be a potent agent against lead acetate-induced testicular toxicity.

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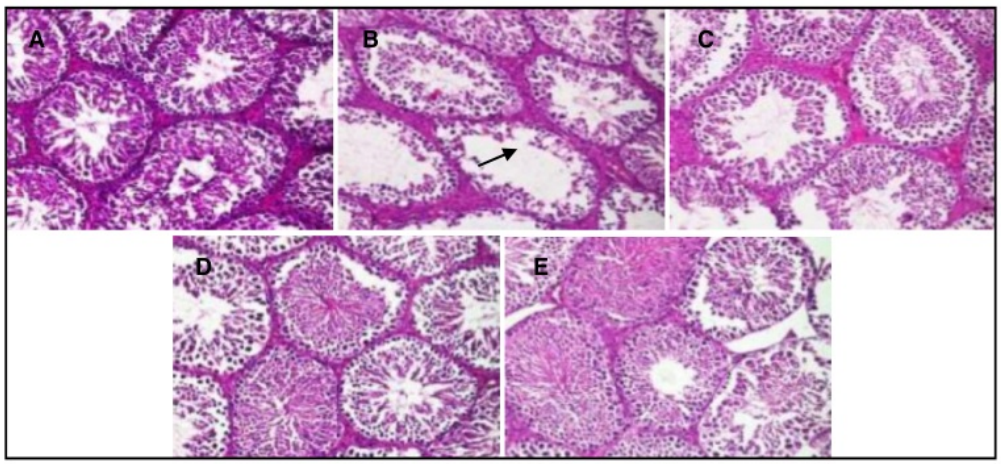

Fig 1: (A) Normal testis in negative control ; (B) The positive control shown seminiferous tubules appear destroyed, loss of spermatogenic cell and necrosis (indicated by arrows); (C and D) Treated showed like a lead acetate treated group; and (E) Treated with 500 mg/kg showed regeneration in testicular cells. (H&E×400).

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Probiotic Utilization in Megacolona Dog : A Case Report

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(Received : April, 2019 **118/19** Accepted : May, 2019)

Abstract

A five year old male beagel dog weighing 15 kg was presented with history of constipation, frequent vomiting after feeding, and abdominal pain for past 3 days to Veterinary Teaching Hospital of Airlangga University, Surabaya. Based on hematological analysis, normal X-ray and contrast X-rayradiography, the dog was diagnosed with megacolon due to hard faeces accumulation in the colon. It was treated with fluid therapy using NaCl, combined with ranitidine, antasids, enrofloxacycin, and probiotic contained *Lactobacillus acidophilus, Bifidobacterium longum, Streptococcus thermophillus* ($1x10^9$ CFU/g per day). This treatment resulted good progress everyday, the dog was totally back

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