Acut Toxicity Test of ImperataCylindrica L. (Beauv) Root Methanol Extract on Mice an a Antihypertention

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Abstract: <u>Purpose</u>: The aim of this study was to evaluate the acut toxicity of methanol extract of Imperatacylindricaroot that at the doses 60 and 90 mg/kg-bw had anti-hypertension effect on rat. <u>Method</u>: OECD guidelines, three groups of mice received oral administration of control solution and methanol extract of Imperatacylindricaroot at the doses285.7; and 714.3mg/kg-bw, respectively. The body weight and the signs of toxicity including mortality and duration of effects were observed. <u>Result</u>: The signs of toxicity which were observed at: 0; $\frac{1}{2}$, 1; and 2 hours, respectively, did notappearfromall groups. Then in the furtherobservations for 14 days did not show the toxic effectand mortality. The body weight of all group increased. Statistically, the difference of weight increasedwas not significant. <u>Conclusion</u>: The methanol extract of Imperatacylindrica root till at the dose 714.3 mg/kg-bw did not cause the toxic effect on mice, and LD₅₀ value suggested higher than 714.3 mg/kg-bw.

Keywords: Acut toxicity, Imperatacylindricaroot methanol extract, Imperatacylindrica

1. Introduction

Prevalence of Hypertensionis high in Indonesia. Many adverse effects were caused by hypertension, and the effects became the problem forpublic health [1]. The Basic of health research (Riskesdas) in 2013, showed that 49% of Indonesian society used herbal medicine to maintain health and to treat the disease [2]. The data shows that jamu as a part of traditional medicine has been accepted by the Indonesian society [3]. It as a traditional medicine is a part of pharmaceutical group, and a part of the resources needed to succeed The Long Term of Development Plan for the Health (in 2005 – 2025) [4].

Traditional medicine (jamu) still needed the sufficient scientific evidence to be used by health professionals. Decree of the Ministerof HealthNumber 760/Menkes/Per/IX/1992 on phytopharmaca, and Number 761/Menkes/SK/IX/1992 onguidelines of phytopharmaca, and Regulation of the Minister of Health number 003/Menkes/Per/2010 on herbs saintification, are some government regulations in an effort to develop of herbal medicinein the formalused together with chemical-based drugs, which provide the scientific evidence related to quality, safety, and the benefits of traditional medicine (herbal medicine) [4-8].

Imperatacylindrica root has been used as a traditional medicine to lower of high blood pressurebysocietyinSouth-East Sulawesi. Ruslinet.al, reported the methanol extract of *Imperatacylindrica* root at doses 60 and 90mg/kg-bw had anti-hypertensi effect on Wistar rat which was measured byTailcuff MP100 PulseTransducer method [9]. The other used in traditional medicine were for fever, jaundice, edema [10]. Parvathy et al.have published that the methanol extract of *Imperatacylindrica* root had anti-anthelmintic activity [11].

Three classess of herbal medicine these are Traditional Medicine, Standardized Herbal Medicine and Phytopharmaca. To promote of using of *Imperatacylindrica* extract as Standardized Herbal Medicine is needed the other tests, one of it toxicity test [12].

The next step which must be done for the safety and efficacy of the use of herbal medicine in human is the toxicity testof the extract using the experimental animals, e.g mice to determine the LD50 and acute toxicity, rat todetermine thesubchronic and chronic toxicities [12,13].

2. Material and Methods

Object: 6 adult female miceDDY strain, weight 29 – 31 g.from School of Pharmacy, Institut Teknologi Bandung.

Sample:

- 1) The maximum dose for rat is 5000 mg/kg-rat. The tested extract for mice $=\frac{1}{7} \times 5000$ mg/kg-bw mice = 714.3 mg/kg-bw mice. It extract was made a suspension/emulsion in 20 ml 0.5% of carboxy methyl cellulose solution
- The control solution is 0.5% CMC solution at the dose 20 ml/kg-bw of mice.

Equipment: the balancesare Sartorius 2442, Scaltec51.

Procedure [14]

The method of the study was approved by the Committee of Animal Ethics of Faculty of Medicine, Universitas Kristen Maranatha – R.S.Immanuel No. 169/KEP/V/2014 on 22 May 2014.

Six mice were divided into two groups, which were 3 mice as control and 3 mice as tested groups, andkept in cages for 5 days for acclimatisation to the laboratory condition.

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- Mice was fasted from food but not water for 4 hours, after that tothe control and the tested groups were given (p.o) with a single dose of control and tested solutions, respectively. The signs of toxicity and duration of effects: motor activity, catalepsy, ptosis, lacrimation, straub tail, piloerection,pinna reflex, and corneal reflex were observed at: 0; ½, 1; 2 hours, respectively.
- 2) Then the signs of toxicity including mortality; nature, severity were observeddaily for 14 days.
- 3) The individual weights of mice were observed at the day of dosing, in weekly intervals thereafter, and at day14.

3. Result

1) The signs of toxicity and duration of effects

 Table 1: The signs of toxicity and duration of effects after oral administration of single dose of Imperatacylindrica root methanol extract

Group Dose (mg/kg bw) and time of observation (hour)												
	Control				Tested							
Observation	0			285.7				714.3				
	0	1⁄2	1	2	0	1⁄2	1	2	0	1⁄2	1	2
Motor activity	Ν	Ν	Ν	Ν	Ν	Ν	Ν	Ν	Ν	Ν	Ν	Ν
Catalepsy	-	-	-	-	-	-	-	-	-	-	-	-
Ptosis	-	-	-	-	-	-	-	-	-	-	-	-
Lacrimation	-	-	-	-	-	-	-	-	-	-	-	-
Straub tail	-	-	-	-	-	-	-	-	-	-	-	-
Piloerection	-	-	-	-	-	-	-	-	-	-	-	-
Pinna reflex	+	+	+	+	+	+	+	+	+	+	+	+
Corneal reflex	+	+	+	+	+	+	+	+	+	+	+	+

Note: N = Normal

- = has not effect

+ = has effect

2) The observation of signs of toxicity including mortality; nature, severity whichwere observed daily for 14 days.

 Table 2: The percentage of mortality of mice during 14 days after oral administration of single dose of Imperatacylindrica

 root methanol extract

root methanol extract							
Groups	The mortality observation						
	during 14	4 days (%)					
	1 - 14	1 - 14					
Control (0 mg/kg-bw)	0	0					
Tested (285.7mg/kg-bw)	0	0					
Tested (714.3 mg/kg-bw)	0	0					

3) The observation of individual weights of mice whichwere observed at the day of dosing, in weekly intervals thereafter, and at day14.

 Table 3: The Body weight at days0 (dosing), 7 and 14 after oral administration of single dose of Imperatacylindrica root methanol extract

	methanor extract										
No.		Body weight of mice (g) at day									
ofmice	Control group (0 mg/kg-bw)					both weight					
	0	7	14	weight	0	7	14	Weight increase	increase		
				increase				-			
1	29.70	32.58	33.30	3.60	29.35	31.72	32.51	3.16	NS		
2	30.22	32.01	33.61	3.39	29.41	31.29	33.84	4.43			
3	30.35	31.56	33.58	3.23	29.30	31.72	32.51	3.21			
Average	30.09 ± 0.26	32.05 ± 0.35	33.50 ± 0.13	$3.41{\pm}~0.13$	29.35 ± 0.04	31.58 ± 0.19	32.95 ± 0.59	3.60 ± 0.55			

Note: NS = Not significant

4. Discussion

The observation fact of the signs of toxicity and duration of effects showedthe all signs of poisoining effect did not appear from all groups. Thus the dose given was not toxic to the groups. Then from the further observations, the facts of the bothgroups did not show the toxic effect and dead.

The body weight the both groups were increase. Statisticaly of the differences of the both increasewere not significant.

Result: The signs of toxicity which were observed at: 0; ¹/₂, 1; and 2 hours, respectively, did notappearfromall groups. Then in the furtherobservations for 14 days did not show the toxic effectand mortality. The body weight of all group increased. Statistically, the difference of weight increased was not significant.

Conclusion: The methanol extract of *Imperatacylindrica* root till at the dose 714.3 mg/kg-bw did not cause the toxic effect on mice, and LD_{50} value suggested higher than 714.3 mg/kg-bw.

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5. Conclusion

The methanol extract of *Imperatacylindrica* root at the dose 714.3 mg/kg-bw did not cause the toxic effect on mice, and the LD_{50} we suggest higher than the dose.

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