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Lethal Dose 50 (LD ^{50}) Extract Ethanol Meat Oyster

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Abstract
The aim of this research was to know Lethal Dose 50 ethanol extract of oyster meat, to know the weight change in mice after giving oyster ethanol extract for seven days. This research method was true experimental by using time series control group design. The test object was 45 male (Mus musculus) mice. They were divided into 9 groups: one control group, and eight treatment groups. Observation of dead animals was done for up to seven days. The results of this study were the ethanol extract of oyster meat did not cause death in animal experiments.Treatment groups that gained weight from baseline body weight were 8%, 12%, 14%, and 16%, and the weight-loss group was in the 32%. The conclusion of this study was LD^{50} ethanol extract of oyster meat was pseudo because it did not cause death in animal try to the highest dose limit of 10.64 g / kgbw. The best weight change was achieved in the 8% mice group.

Keywords: *Oyster, ethanol extract, LD^{50}, mice*

I. INTRODUCTION

One type of animals that live mangrove enforced is an oyster that lives attached to mangrove plants, and these oysters have a high economic value and can be used as food sources of animal protein is high enough. The oyster is a water-dwelling animal (diasis), the upper shell is smaller than the bottom shell. The edge of the shell is not crenul (not crenulated), does not have chomata (denticles). Shell shape is slightly elongated and concaveOysters contain D-aspartic acid (D-Asp) and N-methyl-Daspartate (NMDA) compounds. This compound as the main neurotransmitter and a central nervous stimulant that can trigger the release of GnRH such as Luteinizing Hormone (LH), Follicle-Stimulating Hormone (FSH) and Growth Hormone (GH). These hormones trigger leydig cells in the testes to produce testosterone hormone and maturation of spermatozoa (D'aniello, et al., 2000).

Hasani, et al. (2015) found that there was an influence between giving Meat Ethanol Extracts to Increased Sexual Behavior of Male Mice (introducing p value = 0.002 and mounting, p value = 0.003). Levels of Testosterone Hormone (nmol / L) were higher in the treatment group with a mean value of 31.82 ± 9.00 than in the control group with a mean value of 29.95 ± 3.94. The expression of zinc finger protein (μg/μl) gene in the treatment group was 3.06 ± 6.73 while the control group of mean values was 0.0004 ± 0.0009.

The purpose of this study was to determine the lethal dose of 50 (LD50) of ethanol extract of oyster meat.

II. METHODS

The type of this research was a true experimental study. Determination of sample size according to WHO provisions, with a minimum sample size of 5 for each group. The study materials were oyster, 96% ethanol solution, aquadest, Na CMC. While the equipment used were cage mice, stomach sonde, balance, filter cloth, chemical tube, medium container, pipette, jar, centripuge, rotavapor, and convection oven.

Method of making ethanol extract of oyster meat was the first cleaned after the oyster is done. Oyster meat was further macerated using a 70% ethanol solvent with a material ratio of: 1: 2 solvent (w / v). The maceration process was done three times. The first maseration was done for 3 times 24 hours and the second and third maceration was done for once 24 hours. Maseration results further screening with filter cloth. After the maceration process centrifuge at a speed of 3500 rpm for 60 minutes. The supernatant (liquid phase) was separated from the precipitate (residue). Furthermore, the supernatant was evaporated using a rotary vacuum evaporator at a temperature of 54 ° C for 60 minutes. The resultant rotary evaporator was then fed into the convection oven until the oyster extract was formed powder.

The LD^{50} test was done. Prior to the treatment the mice performed adaptation for 7 days with kept in a cage sized (40x20x5) cm. Each cage contains five mice. The temperature in the cage was set at room temperature, all mice used in the study were fasted for 24 hours before treatment was given. Every day the mice were fed and drinking water was given ad libitum. Then the mice were divided into 9 groups, each group was 5 mice, where the control group was given 1% Na CMC solution and the treatment group was given suspense of ethanol extract of oyster meat with concentration of 1%, 4%, 8%, 10%, 12%, 14%, 16% and 32% orally by using gastric sonde, each group was given different doses according to concentration, then observations were made.

The data have been obtained statistically analyzed by using computer program. The statistical test used is Probit Analysis using Reed and Muench method.

III. RESULTS

Lethal Test Dose 50 (LD^{50})

Testing LD^{50} extract of ethanol oyster with a minimum dose of 0.33 g / kgBWto a maximum dose of 10.66 g / kgBW given orally did not cause death in animal experiments so as not to obtain LD^{50} value of minimal dose to maximal dose as shown in table below this.

Table 1. Number of mice that died and lived after administration of ethanol extract ofoyster meat

Concentrations of dosage% (b/v)	dosage g/kgBW	Number of mice	Number of mice	
			Dead mice	Living mice
Control	0	5	0	5
1% b/v	0.33	5	0	5
4% b/v	1.33	5	0	5
8% b/v	2.66	5	0	5
10% b/v	3.33	5	0	5
12% b/v	4	5	0	5
14% b/v	4.66	5	0	5
16% b/v	5.32	5	0	5
32% b/v	10.64	5	0	5

From table 1 found that the ethanol extract of oyster meat does not cause death effects in animals try mice that have been treated with various concentrations for seven days.

Changes Weight

Table 2. Body weight data of the mice during the trial (starting from day I, III, V, VI and VII)

Group	BW (g)					Weight loss				
	The Day-									
	I	III	V	VI	VII	I-III	III-V	V-VI	VI-VII	VII-I
Control										
1	29.09	28.3	30.9	30.2	30.4	0.79	-2.6	0.7	-0.2	-1.31
2	32.2	30.4	32.9	31.7	30.8	1.8	-2.5	1.2	0.9	1.4
3	30.5	30.4	32.5	32.2	31.5	0.1	-2.1	0.3	0.7	-1
4	27.6	26.3	28.5	28.5	27.8	1.3	-2.2	0	0.7	-0.2
5	28.1	27.2	30.1	29.7	30.3	0.9	-2.9	0.4	-0.6	-2.2
1%										
1	37.4	35.2	35.1	35.6	35.9	2.2	0.1	-0.5	-0.3	1.5
2	32.2	29.8	30.2	31.4	32.1	2.4	-0.4	-1.2	-0.7	0.1
3	24.4	22.9	22.9	32.2	25.7	1.5	0	-9.3	6.5	-1.3
4	34.1	31.1	31	32	33.8	3	0.1	-1	-1.8	0.3
5	38.2	36.3	36	36.2	37.8	1.9	0.3	-0.2	-1.6	0.4
4%										
1	31.4	28.7	31.8	31.3	31.5	2.7	-3.1	0.5	-0.2	-0.1
2	32.9	29.5	31.2	33	32.7	3.4	-1.7	-1.8	0.3	0.2
3	36.9	33.5	37.2	37.4	37.5	3.4	-3.7	-0.2	-0.1	-0.6
4	31.7	29.5	32.2	32.2	32.2	2.2	-2.7	0	0	-0.5
5	30	27.6	30.8	31.9	31.7	2.4	-3.2	-1.1	0.2	-1.7
8%										
1	27.5	30	30.3	28.6	29.6	-2.5	-0.3	1.7	-1	-2.1
2	25	26.6	27.1	26.1	26.6	-1.6	-0.5	1	-0.5	-1.6
3	24	26.2	26.5	25.8	26.8	-2.2	-0.3	0.7	-1	-2.8
4	25.3	26.9	27.9	26.5	27.3	-1.6	-1	1.4	-0.8	-2
5	24.5	27.4	27.6	26.5	27.7	-2.9	-0.2	1.1	-1.2	-3.2
10%										

Group	BW (g) The Day-					Weight loss				
	I	III	V	VI	VII	I-III	III-V	V-VI	VI-VII	VII-I
1	27.6	28.4	27.9	30.1	29.1	-0.8	0.5	-2.2	1	-1.5
2	28.9	28	27.9	30.4	29.9	0.9	0.1	-2.5	0.5	-1
3	27.5	26.6	26.6	27.7	26.3	0.9	0	-1.1	1.4	1.2
4	22.9	22.8	22	24.1	21.7	0.1	0.8	-2.1	2.4	1.2
5	22.6	22.6	23	25.4	24.2	0	-0.4	-2.4	1.2	-1.6
12%										
1	30	31.6	30.6	29.2	31.4	-1.6	1	1.4	-2.2	-1.4
2	27.1	28.9	27	25.5	28.4	-1.8	1.9	1.5	-2.9	-1.3
3	25.3	26.8	26.6	24.3	28.1	-1.5	0.2	2.3	-3.8	-2.8
4	26.9	26.5	26.3	24.2	28.1	0.4	0.2	2.1	-3.9	-1.2
5	35.2	37.2	37.2	33.9	38.2	-2	0	3.3	-4.3	-3
14%										
1	29.9	26.1	29.8	30.4	31.2	3.8	-3.7	-0.6	-0.8	-1.3
2	31.1	28.3	32.3	32.6	32.6	2.8	-4	-0.3	0	-1.5
3	32.7	29.8	33.6	33.2	33.7	2.9	-3.8	0.4	-0.5	-1
4	25.3	22.7	27	27.3	27.6	2.6	-4.3	-0.3	-0.3	-2.3
5	30.8	27.6	31.1	32.3	32.5	3.2	-3.5	-1.2	-0.2	-1.7
16%										
1	27.5	24.2	27.9	27	27.8	3.3	-3.7	0.9	-0.8	-0.3
2	26.4	23.1	27.4	26.8	27.7	3.3	-4.3	0.6	-0.9	-1.3
3	25.6	23.7	26.8	26.6	27.3	1.9	-3.1	0.2	-0.7	-1.7
4	24	20.8	25.1	23.6	24.5	3.2	-4.3	1.5	-0.9	-0.5
5	28.1	23.4	28.9	27.7	29.2	4.7	-5.5	1.2	-1.5	-1.1
32%										
1	26.6	23	21.8	26.2	26	3.6	1.2	-4.4	0.2	0.6
2	36.4	34.5	33.4	35.9	35.2	1.9	1.1	-2.5	0.7	1.2
3	28.9	26.5	26	28.9	28	2.4	0.5	-2.9	0.9	0.9
4	36.4	33.7	33	32.9	30	2.7	0.7	0.1	2.9	6.4
5	39.6	37.4	35.6	38.8	37.9	2.2	1.8	-3.2	0.9	1.7

From the development of body weight obtained there are several treatment groups that all experienced weight gain from the initial body weight of the group of 8%, 12%, 14%, and 16%. There are also groups that all mice lose weight ie 32% group.

IV. DISCUSSION

Based on the observations made during the research, there are several things that need to be discussed are as follows:

The researchers found that none of the mice died after being treated, so the data could not be processed. Based on the agreement taken by the experts, if the maximum dose does not lead to the death of the experimental animal, then the LD^{50} is expressed by LD^{50} 'pseudo' by taking the maximum dose. So in this study LD^{50} known as LD^{50} pseudo, which is 10.64g / KgBW.

This result can not be included in the Doull and Casarett criteria, since the obtained LD^{50} is not a true LD^{50}. The dose of 10.64 g / KgBW is the maximum dose conversion in humans to mice based on the EOCD proposal. Based on the agreement of experts, if at maximum doses there is no death in animals try, then obviously compounds.

Are included in the "Practical No Toxic" criteria so that the maximum dose in humans is converted to 10.64 g / Kg BW in mice, where the dose does not cause death to all experimental animals, including the "Practical No Toxic" criteria in the Doull & Casarett (1986) criteria.The highest dose in this study was 10.64 g / KgBW. While the maximum dose allowed for experiments using mice was 15 g / KgBW, so it has not reached the maximum recommended dose and has not resulted in the mortality of the experimental animals.Researchers did not find any significant abnormalities that occurred in whole-group mice after treatment of qualitative test of toxic symptoms.

From the development of body weight obtained there are several treatment groups that all experienced weight gain from the initial body weight of the group of 8%, 12%, 14%, and 16%. There are also groups that all mice lose weight ie 32% group

V. CONCLUSIONS

1. LD^{50} ethanol extract of oyster meat is a pseudo-LD^{50} at a dose of 10.64g / kgBW because it does not cause death in experimental animals up to the highest dose limit of 10.64 g / kgbw.
2. The best weight change achieved in the mice group 8%

VI. SUGGESTION

There is a need for further research on LD^{50} extract of ethanol oyster using higher doses.

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