ORIGINAL ARTICLE

Chronic toxicity study of *Hyptis suaveolens* (L.) Poit in rats

Aimmanas Attawish[1], Songpol Chivapat[2], Pranee Chavalittumrong[3], Songpol Phadungpat[4], Jaree Bansiddhi[5] and Bunjong Chaorai[6]

Abstract

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Chronic toxicity study of *Hyptis suaveolens* (L.) Poit in rats
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The effect of water extract of *Hyptis suaveolens* (*H. suaveolens*) was evaluated for 6-month chronic toxicity in Wistar rats. Control group received distilled water orally 10 ml/kg/day. The extract was orally given to five treatment groups at the doses of 5, 50, 250, 500 and 500 mg/kg/day for 6 months. The last group was served as the recovery group. Changes in the body weights, actual and relative organ weights were not significantly demonstrated in all groups throughout the study. The results of hematological, biochemical parameters and histopathological lesions showed that the extract did not produce any significant dose-related changes. Therefore, it may be concluded that the extract of *H. suaveolens* at the given doses did not produce any significant toxic effect in rats during 6-month period of the treatment.

Key words : *Hyptis suaveolens*, chronic toxicity, rat

[1]M.Sc.(Nutritional Toxicology) [2]M.Sc.(Pathobiology), D.V.M. [3]M.S.(Phytochemistry), B.Sc. in Pharm. [4]B.Sc. (Biotechnology) [5]M.Sc.(Botany) [6]B.Sc.(Botany), Medicinal Plant Research Institute, Department of Medical Sciences, Ministry of Public Health, Nonthaburi 11000, Thailand.
Corresponding e-mail: aimmanas@dmsc.moph.go.th
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บทคัดย่อ

เอมมนัส อัตตวิชญ์ ทรงพล ชีวะพัฒน์ ปราณี ชวลิตธำรง ทรงพล ผดุงพัฒน์ จารีย์ บันสิทธิ์ และ บรรจง ชาวไร่

การศึกษาพิษเรื้อรังของแมงลักคาในหนูขาว

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ประเมินผลการศึกษาพิษเรื้อรังของสารสกัดด้วยน้ำของแมงลักคาในหนูขาวเป็นระยะเวลานาน 6 เดือน เปรียบเทียบกับหนูกลุ่มควบคุมที่ได้รับน้ำกลั่นขนาด 10 มล./กก./วัน โดยป้อนสารสกัดให้หนูกลุ่มทดลอง 5 กลุ่ม ในขนาด 5, 50, 250, 500 และ 500 มก./กก./วัน โดยหนูกลุ่มสุดท้ายเป็นหนูกลุ่มสังเกตอาการหลังการหยุดยา การเปลี่ยนแปลงของน้ำหนักตัวหนูขาว และน้ำหนักอวัยวะในหนูกลุ่มทดลองทุกกลุ่มไม่แตกต่างอย่างมีนัยสำคัญจากกลุ่มควบคุม ผลการศึกษาทางโลหิตวิทยา ชีวเคมีคลินิก และจุลพยาธิวิทยา ไม่พบการเปลี่ยนแปลงที่แตกต่างอย่างมีนัยสำคัญในหนูขาวที่สัมพันธ์กับขนาดของสารสกัดที่ได้รับ ดังนั้นอาจสรุปได้ว่าสารสกัดแมงลักคาในขนาดที่ให้หนูขาวไม่ก่อให้เกิดอาการพิษตลอดระยะเวลา 6 เดือนที่ทำการศึกษา

สถาบันวิจัยสมุนไพร กรมวิทยาศาสตร์การแพทย์ กระทรวงสาธารณสุข จังหวัดนนทบุรี 11000

Hyptis suaveolens (L.) Poit (*H. suaveolens*) (Labiatae) is commonly called in Thai as "Maeng lak kha" (Smitinand, 2001). It is a strong-scented herb, 30-150 cm high; spreading stem hispid. Leaves are ovate to broadly ovate, 2-8 cm long, 2-6 cm wide, pubescent; and have serrulate margins. Flowers are arranged in vertcillate cymes, calyx campanulate, 5-toothed, spine-like teeth; corolla, blue or bluish violet, 2-lipped. Fruitlets are flattened, broadly obovoidal and emarginated at apex; pericarp swelling to a gelatinous mass when soaked in water (Keng, 1978; Li and Hedge, 1994). The plant is naturalized as weeds in open areas, found throughout Thailand. The main chemical constituents were 1,8-cineole and β-caryophyllene obtained from distillation (Peerzada, 1997). Azevedo *et al*. (2001) reported that the essential oils of *H. suaveolens* were sabinene, limonene, bicyclogermacrene, β-phellandrene and 1,8-cineole. After that Ziegler *et al*. (2002) isolated dehydroabietinol from this plant. *H. suaveolens* was reported to be of therapeutic value as a stimulant carminative, antiseptic, sudorific and galactagogue (Saluja and Santani, 1993). The essential oil of *H. suaveolens* inhibited the growth of both gram-positive and gram-negative bacteria as well as had mild antifungal activity against *Candida albicans* and *Aspergillus niger* (Iwu *et al*., 1990). Ethanolic extract of *H. suaveolens* leaves showed wound healing activity. The activity may be due to free radical scavenging action of the plant and enhancing level of antioxidant enzymes in granuloma tissue (Shirwaikar *et al*., 2003).

However, no report for long-term toxicity test has been made. Thus in order to obtain safety data of this plant extract prior to the clinical trial of *H. suaveolens* in humans, six-month toxicity study of *H. suaveolens* water extract was performed in rats.

Materials and Methods

Plant material

The aerial part of *H. suaveolens* was collected from Kanchanaburi Province. The botanical identifications were determined using description of Keng (1978) and Li and Hedge (1994), and compared with the authentic specimen (Kerr10204) at the Bangkok Herbarium (BK), Department of Agriculture, Ministry of Agriculture and Cooperative, Bangkok, Thailand. A voucher specimen (Bansiddhi 45-1) was deposited at the Botanical Laboratory, Medicinal Plant Research Institute, Nonthaburi, Thailand.

Plant extraction

H. suaveolens aerial part, was washed with water, cut and dried in an oven at 50ºC then refluxed. The filtrate was dried to give a residue (yield: 13.0 % w/w). The residue was dissolved in distilled water to concentrations needed for the study.

Animals

Ninety male Wistar rats weighing 150±10 g and 90 female rats weighing 130±10 g from the National Laboratory Animal Center, Mahidol University, Nakornpathom province, were used. The animals were housed in the animal facility of the Department of Medical Sciences, Ministry of Public Health, maintained in standard environmental conditions and were allowed to have free access to food and clean water.

Six-month toxicity study

According to the WHO guideline (2000), ninety Wistar rats of each sex were randomly divided into 6 groups of 15 animals per sex. Group 1 (water control) received distilled water 10 ml/kg/day for 6 months. Groups 2-5 were orally treated with the water extract at the doses of 5, 50, 250 and 500 mg/kg/day which were equivalent to 1, 10, 50 and 100 folds the therapeutic dose in human, respectively. A further group, group 6, was treated with 500 mg/kg/day and used as a recovery group. Body weight and food-intake were measured weekly and the animals were observed for signs of abnormalities during the six-month of the treatment. At the end of the treatment period, the 1^{st}-5^{th} groups of rats were fasted for 18 hours, then anesthetized with ether and sacrificed by drawing blood samples from the posterior vena cava for hematological and biochemical examinations. The 6^{th} group of the rats, the recovery group, was withdrawn from of the feeding of the extract for 14 days before being sacrificed in order to determine whether toxic effects observed in high-dose group were reversible or permanent.

Hematological analysis was performed using an automatic hematological analyzer (Cell Dyn 3500, Abbott). The parameters of the blood samples measured were: hematocrit (Hct), hemoglobin (Hb), red blood cells (RBC), mean cell volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), white blood cells (WBC), % neutrophil (%N), % lymphocyte (%L), % monocyte (%M), % eosinophil (%E), % basophil (%B) and platelet.

Biochemical analysis of serum samples was performed using an automatic chemistry analyzer (Hitachi Model 912, Roche). Biochemical parameters measured were alkaline phosphatase (ALP), alanine aminotransferase (ALT), aspartate aminotransferase (AST), blood urea nitrogen (BUN), total protein, albumin, bilirubin, creatinine, glucose, uric acid, triglyceride, cholesterol, sodium, potassium and chloride.

The positions, shapes, sizes and colors of the internal organs; namely, brain, heart, both kidneys, lungs, salivary gland, thyroid gland, parathyroid gland, trachea, esophagus, stomach, liver, pancreas, intestine, spleen, bladder, adrenal gland and testis in male rats or ovary and uterus in female rats were visually observed for any signs of gross lesions. These organs were then collected, weighed to determine relative organ weights, and preserved in 10% phosphate buffered formalin solution. Tissue slides were prepared and stained with hematoxylin and eosin. Histopathological examination was performed by a pathologist.

Statistical Analysis

The data were analyzed by one-way ANOVA followed by Dunnett multiple ranges test to determine significant differences between groups at $P<0.05$. Histopathological data were evaluated by the Fisher exact test and the significance level was set at $P<0.05$.

Results and Discussion

Effects of *H. suaveolens* extract on body weight, food-intake and relative organ weight

In both male and female rats, there was no difference in the average body weights between extract-treated groups and control groups through-

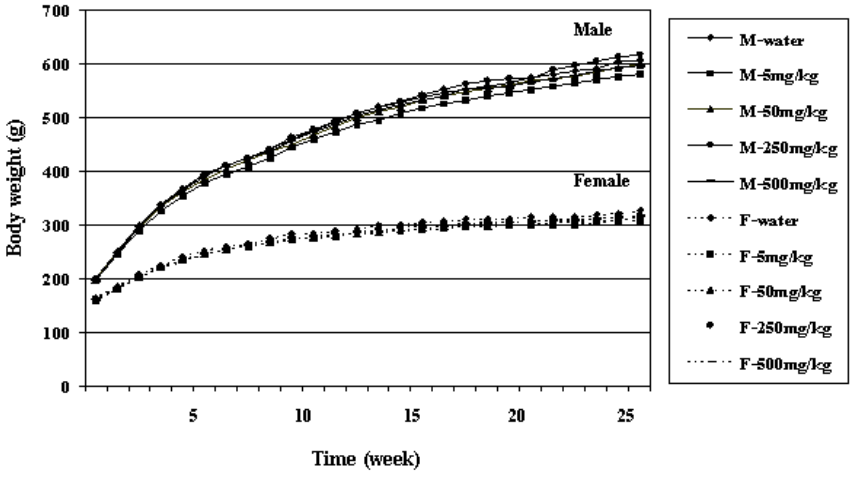

Figure 1. Growth curves of male and female rats receiving *H. suaveolens* for 6 months.

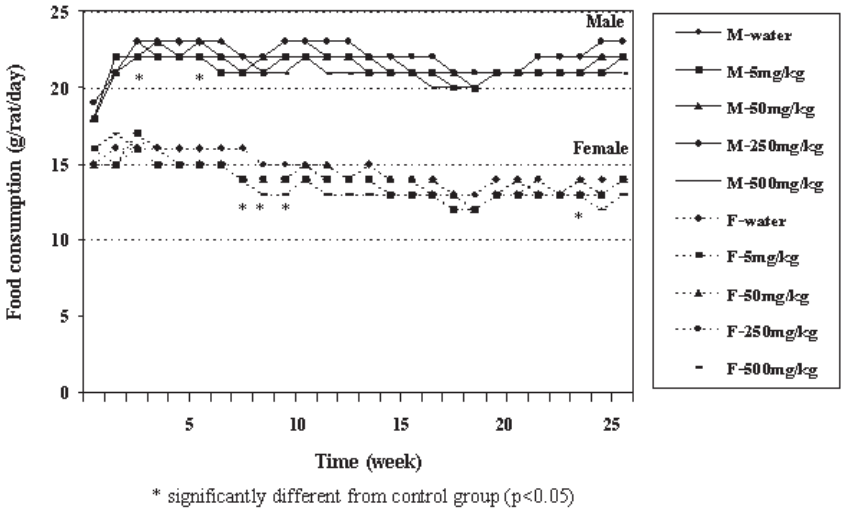

Figure 2. Food consumption of male and female rats receiving *H. suaveolens* for 6 months.

out the experimental period of six months (Figure 1). However, relative food consumption of rats receiving the extract was significantly different from the control groups for several weeks. Male rats receiving 5 mg/kg/day and 5, 50 mg/kg/day of the extract had significantly lower relative food-intake than the control group during 3rd and 6th weeks, respectively. All female rats receiving the same amount of extract as male rats also had significantly lower relative food-intake than the control group at 8th week. During 9th, 10th and 24th weeks they showed the same incidence at the dose of 500 mg/kg/day (Figure 2). In both male and female rats, there was no difference in the relative organ weight between extract-treated groups and control groups (Tables 1 and 2).

Effect of *H. suaveolens* on hematological parameters

There was no difference of the number of red blood cells, hemoglobin, hematocrit, MCV, MCH, MCHC, the number of white blood cells,

Table 1. Body weight (g) and relative organ weight (g/kg) of male rats receiving *H. suaveolens* for 6 months.

Organs	Dose of *H. suaveolens* (mg/kg BW/day)					
	control	5	50	250	500	500-R
	n = 15	n = 15	n = 15	n = 15	n = 15	n = 15
Initial body weight	151±9	146±9	147±7	146±9	147±8	149±10
Final body weight	606±49	581±38	599±56	618±48	596±60	586±60
Brain	3.74±0.34	3.72±0.32	3.65±0.42	3.50±0.37	3.70±0.33	3.73±0.40
Heart	2.48±0.25	2.40±0.21	2.40±0.20	2.36±0.18	2.37±0.22	2.58±0.24
Lung	3.08±0.41	3.03±0.33	3.06±0.25	2.93±0.25	3.00±0.31	2.82±0.61
Liver	24.43±1.71	23.70±1.30	23.98±1.71	23.55±1.49	23.81±1.32	24.42±1.51
Stomach	3.79±0.49	3.59±0.36	3.82±0.43	3.56±0.45	3.69±0.36	3.74±0.40
Spleen	1.71±0.33	1.71±0.25	1.78±0.50	1.60±0.17	1.67±0.24	1.70±0.20
Right Kidney	2.28±0.20	2.23±0.20	2.28±0.20	2.16±0.21	2.27±0.25	2.31±0.21
Left Kidney	2.18±0.19	2.23±0.20	2.21±0.23	2.09±0.19	2.21±0.20	2.25±0.18
Right Testis	5.50±0.68	5.47±0.71	5.08±0.60	5.22±0.52	5.29±0.56	5.47±0.66
Left Testis	5.47±0.58	5.47±0.68	5.19±0.63	5.23±0.57	5.40±0.62	5.56±0.63
Right Adrenal	0.058±0.018	0.062±0.020	0.055±0.012	0.057±0.008	0.061±0.013	0.057±0.013
Left Adrenal	0.065±0.020	0.064±0.021	0.060±0.014	0.063±0.010	0.067±0.011	0.065±0.015
Bladder	0.287±0.076	0.268±0.048	0.242±0.055	0.261±0.068	0.278±0.065	0.303±0.055

R: Recovery group
The values are expressed as mean ± SD.

Table 2. Body weight (g) and relative organ weight (g/kg) of female rats receiving *H. suaveolens* for 6 months.

Organs	Dose of *H. suaveolens* (mg/kg BW/day)					
	control	5	50	250	500	500-R
	n = 15	n = 15	n = 15	n = 15	n = 15	n = 15
Initial body weight	137±10	134±10	137±13	135±10	134±12	138±11
Final body weight	327±20	314±31	309±23	318±44	318±27	327±36
Brain	6.32±0.43	6.51±0.77	6.61±0.58	6.30±0.72	6.45±0.52	6.27±0.56
Heart	2.78±0.25	2.86±0.31	3.00±0.28	2.85±0.30	2.85±0.28	3.04±0.34
Lung	4.20±0.43	4.34±0.39	4.41±0.38	4.04±0.49	4.16±0.45	4.14±0.38
Liver	23.64±1.99	24.45±3.25	25.22±2.23	22.70±1.68	23.80±2.37	24.79±1.44
Stomach	5.08±0.43	5.13±0.72	5.27±0.76	4.80±0.58	5.09±0.61	5.09±0.44
Spleen	2.35±0.37	2.27±0.31	2.50±0.52	2.18±0.28	2.25±0.32	2.32±0.32
Right Kidney	2.69±0.24	2.72±0.25	2.73±0.20	2.70±0.28	2.62±0.24	2.76±0.24
Left Kidney	2.54±0.24	2.56±0.23	2.63±0.23	2.55±0.30	2.51±0.23	2.66±0.26
Right Adrenal	0.124±0.020	0.127±0.030	0.136±0.030	0.125±0.024	0.122±0.016	0.131±0.025
Left Adrenal	0.134±0.018	0.138±0.030	0.143±0.021	0.130±0.022	0.125±0.057	0.145±0.024
Bladder	0.278±0.054	0.265±0.036	0.298±0.051	0.289±0.055	0.268±0.038	0.296±0.043

R: Recovery group
The values are expressed as mean ± SD.

Table 3. Hematological values of male rats receiving *H. suaveolens* for 6 months.

Parameters	Dose of *H. suaveolens* (mg/kg BW/day)					
	control	5	50	250	500	500-R
	n = 15	n = 15	n = 15	n = 15	n = 15	n = 15
Hematocrit (%)	49.95±3.51	50.03±1.56	50.29±2.57	50.78±2.55	51.91±1.55	50.40±2.21
RBC ($\times10^6$ cells/mm^3)	9.21±1.00	9.42±0.43	9.36±0.50	9.38±0.64	9.61±0.46	9.33±0.46
Hemoglobin (g/dl)	16.29±1.18	16.29±0.55	16.44±0.71	16.58±0.75	16.99±0.49	15.94±2.16
MCV (μm^3/red cell)	54.51±3.12	53.17±1.46	53.74±1.56	54.21±1.41	54.06±1.80	54.03±1.67
MCH (pg/red cell)	17.77±0.94	17.32±0.43	17.59±0.47	17.70±0.55	17.69±0.53	17.58±0.57
MCHC (g/dl RBC)	32.06±0.45	32.57±0.46	32.70±0.59	32.65±0.47	32.74±0.40	32.53±0.49
WBC ($\times10^3$ cells/mm^3)	5.00±1.43	4.65±0.98	4.68±1.08	4.87±1.67	4.56±0.78	4.67±1.14
Neutrophil (%)	18.26±7.74	14.87±3.44	16.79±4.31	18.38±7.60	17.87±6.67	14.86±3.35
Eosinophil (%)	1.43±0.54	1.55±0.40	1.38±0.56	1.47±0.50	2.13±2.43	1.40±0.38
Lymphocyte (%)	65.97±9.90	71.34±6.15	69.60±5.34	68.02±9.55	67.97±7.78	71.14±4.88
Monocyte (%)	8.81±2.76	7.78±3.36	7.43±2.89	7.74±4.39	8.01±2.33	7.34±2.59
Basophil (%)	5.53±1.93	4.46±1.63	4.80±1.59	4.38±1.68	5.25±2.78	5.26±2.03
Platelet ($\times10^3$ cells/mm^3)	1070±172	1007±106	977±134	952±150	1024±108	1023±109

R: Recovery group
The values are expressed as mean ± SD.

Table 4. Hematological values of female rats receiving *H. suaveolens* for 6 months.

Parameters	Dose of *H. suaveolens* (mg/kg BW/day)					
	control	5	50	250	500	500-R
	n = 15	n = 15	n = 15	n = 15	n = 15	n = 15
Hematocrit (%)	49.59±1.91	51.13±2.77	50.64±2.28	50.16±1.26	51.09±2.39	50.46±2.62
RBC ($\times10^6$ cells/mm^3)	8.49±0.44	8.70±0.49	8.75±0.41	8.64±0.31	8.72±0.50	8.50±0.41
Hemoglobin (g/dl)	16.15±0.55	16.72±0.86	16.56±0.67	16.36±0.40	16.56±0.74	16.32±0.88
MCV (μm^3/red cell)	58.51±1.86	58.77±1.60	57.93±2.21	58.05±1.61	58.65±1.12	59.37±1.63
MCH (pg/red cell)	19.67±0.63	19.23±0.51	18.95±0.64	18.94±0.47	19.02±0.50	19.21±0.62
MCHC (g/dl RBC)	32.57±0.37	32.73±0.54	32.72±0.46	32.64±0.32	32.42±0.34	32.36±0.71
WBC ($\times10^3$ cells/mm^3)	2.12±0.46	2.15±0.55	2.33±0.46	2.18±0.75	2.27±0.53	2.42±0.69
Neutrophil (%)	16.88±3.96	18.12±3.81	18.39±4.58	18.26±5.62	15.74±5.41	14.51±4.46
Eosinophil (%)	1.51±0.46	1.62±0.56	1.57±0.58	1.60±0.58	1.56±0.56	1.40±0.73
Lymphocyte (%)	67.71±7.59	65.22±4.54	64.90±6.21	65.33±9.11	66.68±9.10	62.05±12.01
Monocyte (%)	10.50±5.35	11.01±3.72	10.70±4.67	10.65±4.59	11.48±5.98	16.32±8.82*
Basophil (%)	3.41±1.27	4.03±1.53	4.44±2.13	4.16±2.04	4.19±1.68	5.72±3.01*
Platelet ($\times10^3$ cells/mm^3)	942±79	922±106	959±95	956±120	993±103	950±72

*** significantly different from control group ($p<0.05$)**
R: Recovery group
The values are expressed as mean ± SD.

%N, %L, platelet between extract-treated groups and control groups of either male or female rats (Tables 3 and 4). Whereas in female rats, %M and %B of the recovery group were significantly higher than those of the control group, %M and %B of other extract-treated groups were not different from

those of the control. Hence, these changes may not be due to the effect of the extract.

Effect of *H. suaveolens* on biochemical parameters

In both male and female rats, no differences (Tables 5 and 6) in the serum levels of ALP, ALT, AST, total protein, albumin, bilirubin, creatinine, triglyceride, cholesterol, sodium and potassium were found between *H. suaveolens*-treated groups and the control groups. It was found that serum BUN of male rats receiving *H. suaveolens* at the dose of 500 mg/kg/day was significantly lower than that of the control group; however, the value was still within normal range (Semler *et al*., 1992). Serum glucose of male rats receiving *H. suaveolens* at the dose of 50 mg/kg/day was significantly lower than that of the control group. Since this change did not occur in other *H. suaveolens*-treated groups, it was not a dose-dependent change and should not be due to the plant extract. Male rats receiving *H. suaveolens* at the dose of 250 mg/kg/day and the recovery group had significantly higher and lower levels of chloride ion compared to the control group, respectively; however, the values were within normal range (100-110 mmol/l) (Semler *et al*., 1992) and the changes were not dose-dependent. In female rats, the group receiving *H. suaveolens* at the dose of 500 mg/kg had significantly higher serum uric acid than the control group but the value was still within normal range (1.2-7.5 mg/dl) (Semler *et al*., 1992).

Effect of *H. suaveolens* on histopathology of internal organs

No abnormal signs of internal organs were observed by gross examination. Histopathological examinations of the brain, lung, salivary gland, thyroid gland, parathyroid gland, trachea, esophagus, liver, heart, spleen, pancreas, kidney, stomach, intestine, bladder, including testis and prostate gland in male rats or vagina, cervix, uterus and ovary in female rats were performed. Pathological changes were observed in some tissue samples in some groups of animals. Fatty changes of the liver, myocarditis of the heart, nephrocalcinosis, pyelonephritis and hydrocalyx of the kidney, splenomegaly and spermatic granuloma were found (Tables 7 and 8). However, all of these histopathological findings occurred in a small number of animals and were neither a significant change nor dose-related, and some changes occurred in the control group only.

The result indicated that the water extract of *H. suaveolens* given orally at doses of 5, 50, 250 and 500 mg/kg/day did not produce any sign of toxicity in the rats during the 6-month administration period. *H. suaveolens* was a Thai medicinal plant that was tested for anti-HIV activity in the "Project Herbs for AIDS" of the Department of Medical Sciences, Ministry of Public Health. It was found that water extract of this plant possessed in vitro anti-HIV activity. Using enzyme inhibition assay, it was found that the mechanism of action of this plant extract might be due to the inhibition of HIV-1 reverse transcriptase and protease. Chronic toxicity study of this plant extract was therefore performed in rats in order to obtain safety data of this extract prior to the evaluation of its therapeutic efficacy in humans.

Conclusion

Six-month chronic toxicity study of *H. suaveolens* in Wistar rats indicated that the water extract of *H. suaveolens* at the doses of 5, 50, 250 and 500 mg/kg/day, which were equivalent to 1, 10, 50 and 100 folds the therapeutic dose, respectively, did not produce any significant dose-related changes of hematological parameters, serum biochemistry or histopathology of any internal organs. Therefore, it is concluded that the water extract of *H. suaveolens* at the given doses does not produce any significant toxic effect in rats during a six-month treatment period.

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Table 5. Biochemical values of male rats receiving *H. suaveolens* for 6 months.

Parameters	Dose of *H. suaveolens* (mg/kg BW/day)					
	control	5	50	250	500	500-R
	n = 15	n = 15	n = 15	n = 15	n = 15	n = 15
ALP (U/L)	60.60±10.67	59.13±10.96	58.73±9.35	63.60±10.89	61.00±15.60	60.27±9.97
ALT (U/L)	33.00±8.94	30.13±2.85	30.73±3.90	29.67±4.29	34.27±6.12	30.87±3.98
AST (U/L)	81.00±14.76	82.33±7.22	81.40±9.69	76.60±7.96	80.67±9.98	83.93±8.03
Total protein (g/dl)	6.67±0.38	6.59±0.23	6.74±0.33	6.76±0.25	6.79±0.33	6.71±0.21
Albumin (g/dl)	4.21±0.42	4.26±0.17	4.32±0.18	4.28±0.24	4.41±0.14	4.34±0.09
Bilirubin (mg/dl)	0.08±0.03	0.09±0.03	0.08±0.03	0.08±0.04	0.10±0.04	0.08±0.04
BUN (mg/dl)	19.64±1.84	18.50±1.41	19.00±2.25	18.56±1.67	17.73±2.41*	18.19±2.75
Creatinine (mg/dl)	0.72±0.06	0.73±0.06	0.72±0.06	0.73±0.04	0.75±0.05	0.72±0.04
Glucose (mg/dl)	216.06±49.73	190.04±27.21	179.06±28.33*	200.14±30.71	206.16±33.71	202.84±56.76
Uric acid (mg/dl)	3.98±1.75	3.03±1.71	2.77±1.75	3.06±1.88	3.61±1.88	3.77±1.90
Triglyceride (mg/dl)	103.72±29.00	99.11±27.65	110.42±28.19	111.67±31.59	117.32±28.18	112.84±35.42
Cholesterol (mg/dl)	87.83±14.58	80.97±12.42	84.68±18.12	85.53±20.07	78.98±15.39	80.84±11.18
Na^+ (mmol/l)	145±2	145±2	146±3	146±3	147±3	144±1
K^+ (mmol/l)	6.17±0.98	6.10±1.57	5.70±1.63	5.74±1.05	5.70±0.99	6.02±1.16
Cl^- (mmol/l)	105±2	105±1	106±2	107±1*	106±2	103±1*

*** significantly different from control group ($p<0.05$)**
R: Recovery group
The values are expressed as mean ± SD.

Table 6. Biochemical values of female rats receiving *H. suaveolens* for 6 months.

Parameters	Dose of *H. suaveolens* (mg/kg BW/day)					
	control	5	50	250	500	500-R
	n = 15	n = 15	n = 15	n = 15	n = 15	n = 15
ALP (U/L)	23.27±4.30	24.60±5.67	22.27±5.19	23.60±4.12	23.53±3.81	22.60±4.94
ALT (U/L)	26.67±7.28	33.53±16.87	32.00±12.82	31.07±10.21	31.53±9.19	25.47±6.84
AST (U/L)	84.33±13.99	97.13±29.39	96.60±21.16	94.27±18.99	88.67±11.11	83.67±15.96
Total protein (g/dl)	6.85±0.41	6.92±0.37	6.93±0.34	6.83±0.46	7.07±0.34	7.01±0.29
Albumin (g/dl)	4.85±0.30	4.89±0.20	4.88±0.24	4.87±0.33	5.10±0.30	4.93±0.21
Bilirubin (mg/dl)	0.13±0.04	0.13±0.04	0.12±0.03	0.14±0.06	0.13±0.03	0.11±0.04
BUN (mg/dl)	25.35±4.07	22.13±2.49	23.51±2.43	24.40±3.56	24.65±5.13	21.29±3.48
Creatinine (mg/dl)	0.87±0.10	0.80±0.07	0.84±0.09	0.88±0.12	0.87±0.12	0.87±0.06
Glucose (mg/dl)	125.00±19.95	120.27±28.32	116.69±26.06	127.27±27.42	138.90±32.45	138.33±27.30
Uric acid (mg/dl)	2.07±0.89	2.74±1.18	2.60±1.23	2.64±1.05	3.51±1.37*	3.00±0.91
Triglyceride (mg/dl)	52.16±21.83	49.10±15.86	49.14±14.74	51.30±17.38	62.58±17.60	45.94±5.05
Cholesterol (mg/dl)	68.56±18.32	73.32±17.01	67.98±15.28	60.69±12.76	75.74±17.98	72.21±10.54
Na^+ (mmol/l)	145±2	145±1	145±2	146±2	145±2	145±1
K^+ (mmol/l)	5.17±1.34	6.32±1.36	6.02±1.48	5.53±1.17	6.43±1.47	6.05±1.61
Cl^- (mmol/l)	108±2	109±3	109±2	109±2	109±2	107±1

*** significantly different from control group ($p<0.05$)**
R: Recovery group
The values are expressed as mean ± SD.

Table 7. Histopathological values of male rats receiving *H. suaveolens* for 6 months.

Organs	Microscopic findings	Dose of *H. suaveolens* (mg/kg BW/day)					
		control	5	50	250	500	500-R
		n = 15	n = 15	n = 15	n = 15	n = 15	n = 15
Lung		0/15	0/15	0/15	0/15	0/15	0/15
Heart	Myocarditis	1/15	0/15	0/15	3/15	0/15	0/15
Liver	Fatty change	1/15	0/15	0/15	2/15	0/15	0/15
Kidney	Hydrocalyx	1/15	0/15	0/15	0/15	0/15	0/15
Spleen	Splenomegaly	1/15	0/15	0/15	0/15	0/15	0/15
Pancreas		0/15	0/15	0/15	0/15	0/15	0/15
GI tract		0/15	0/15	0/15	0/15	0/15	0/15
Testis	Spermatic granuloma	0/15	0/15	1/15	0/15	0/15	0/15
Adrenal grand		0/15	0/15	0/15	0/15	0/15	0/15
Salivary grand		0/15	0/15	0/15	0/15	0/15	0/15

R: Recovery group
The results are expressed as number of rats with pathological findings / total number of rats examined.

Table 8. Histopathological values of female rats receiving *H. suaveolens* for 6 months.

Organs	Microscopic findings	Dose of *H. suaveolens* (mg/kg BW/day)					
		control	5	50	250	500	500-R
		n = 15	n = 15	n = 15	n = 15	n = 15	n = 15
Lung		0/15	0/15	0/15	0/15	0/15	0/15
Heart		0/15	0/15	0/15	0/15	0/15	0/15
Liver	Fatty change	0/15	0/15	1/15	0/15	0/15	0/15
Kidney	Nephrocalcinosis	1/15	0/15	0/15	0/15	0/15	1/15
	Pyelonephritis and hydrocalyx	0/15	0/15	0/15	1/15	0/15	0/15
Spleen		0/15	0/15	0/15	0/15	0/15	0/15
Pancreas		0/15	0/15	0/15	0/15	0/15	0/15
GI tract		0/15	0/15	0/15	0/15	0/15	0/15
Ovary		0/15	0/15	0/15	0/15	0/15	0/15
Uterus		0/15	0/15	0/15	0/15	0/15	0/15
Cervix		0/15	0/15	0/15	0/15	0/15	0/15
Adrenalgrand		0/15	0/15	0/15	0/15	0/15	0/15
Salivary grand		0/15	0/15	0/15	0/15	0/15	0/15

R: Recovery group
The results are expressed as number of rats with pathological findings / total number of rats examined.

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