RESEARCH ARTICLE | FEBRUARY 28 2019

Effect of kangxuan granule on long-term toxicity in rats FREE

Wenxian Lu; Jianhua Sun; Chongxian Li; Junjie Ma ✉; Dianjin Wen; Hailong Sun; Youling Huang; Zhengchen Zhang; Yueyan Yu

AIP Conf. Proc. 2079, 020034 (2019)
https://doi.org/10.1063/1.5092412

Effect of Kangxuan Granule on Long-Term Toxicity in Rats

Wenxian Lu [1], Jianhua Sun [1], Chongxian Li [1], Junjie Ma [1, a)], Dianjin Wen [1], Hailong Sun [1], Youling Huang [1], Zhengchen Zhang [1] and Yueyan Yu [2]

[1]*The 371st Central Hospital of the Chinese people's Liberation Army, Xinxiang 453000, China*
[2]*The 155th Hospital of the Chinese people's Liberation Army, Kaifeng 475000, China*

[a)]Corresponding author: majunjie371@126.com

Abstract. To observe the toxicity and extent of long-term oral Kangxuan granules in rats to determine the safety of clinical drug use. Kangxuan granules were treated with 25g kg^{-1} d^{-1} for 12 weeks. The changes of physiologic and biochemical indexes such as appearance, body mass, liver and kidney function, blood glucose metabolism were observed. There was no obvious change in appearance and body mass, and the viscera specific mass and pathological examination were normal. Kangxuan granules in clinical dosage of 50 times the case, rats are safe and non-toxic.

INTRODUCTION

Kangxuan granules are composed of hookifera, Pueraria lobata, Ling Magnet, Salvia miltiorrhiza, Radix Achyranthes bidentata, Ginger Pinellia and Ligusticum chuanxiong. This preparation calms the liver latent yang, invigorates the blood circulation to clear the orifices, exhilarates the wind to fix the dizziness. Due to hypertension, cerebral arteriosclerosis, vertebrobasilar artery insufficiency, Mernier's disease, vertigo caused by autonomic nerve dysfunction, head, foot, walking instability and so on. In order to ensure the safety of clinical drug use, the long-term toxicity test in rats was studied.

MATERIAL

C8000 biochemical automatic Analyzer, American Abbott LH750 Blood Analyzer, Beckman Kurt Company, USA; Kangxuan granules (PLA 371st Hospital preparation Room, batch 20000310); precise weighing of a certain amount of capsule powder, 0.5%CMC-Na was used to prepare suspension with a mass concentration of 2.5~0.5 g m~($^{-1}$), 60 SD rats of 6 weeks of age, with body weight (93.5 ± 6.5) g, half male and female (Hubei Experimental Animal Center, Animal Certificate No. 19-008), experimental rats were reared. In the plastic feed box, the feed is the all-price pure rat pellet feed, the drinking water is the tap water, the laboratory temperature is 20 ~ 20°C, the relative humidity is 60~70.

METHODS AND RESULTS

Methods

SD rats were randomly divided into 3 groups with 20 rats in each group [1-2]. One week before administration, there was no abnormality in animal activity, food intake and feces in each group. (1) High dose group: 25 g kg-1 d-1 (50 times); (2 of human dosage), low dose group: 5 g kg^{-1} d^{-1}, 5 g kg^{-1} d^{-1}, 5 g kg^{-1} d 1, 5 g kg^{-1} d 1, respectively. The control group was given equal volume of 0.5%CMC-Na 10 m 1 kg^{-1} d^{-1} (equivalent to 10 times the human dosage of); (3). Once a day by gavage for 12 consecutive weeks Observe the activity, feeding, feces, hair and so on of rats, weigh

2018 International Conference on Biotechnology and Bioengineering (8th ICBB)
AIP Conf. Proc. 2079, 020034-1–020034-3; https://doi.org/10.1063/1.5092412
Published by AIP Publishing. 978-0-7354-1807-3/$30.00

once a week, if found dead or near death of rats, timely autopsy. Weigh the body weight once a week and adjust the dosage according to the weekly body mass.

Determination of hemoglobin (Hb), platelet number (PLT), red blood cell number (RBC) leukocyte count (WBC) and its classification (DC). 24 hours after the last administration of the drug: hemoglobin (Hb), platelet number (PLT), red blood cell number (WBC) and its classification (DC).

Liver and kidney function were measured. These include: AST, alt, TP, ALB, ALB, AlPU, creabun, GLU, T-CH0. The main organs were examined by autopsy. Brain, heart, liver, spleen, lung, kidney, thymus, thyroid, adrenal gland, prostate, ovary, uterus, testis and epididymis were observed. Weighing in which substantive organ quality and large Rat mass. Calculate the organ coefficient.

Histopathological examination and reversible observation were performed. If abnormal lesions were found in the high dose group, pathological examination should also be carried out in the low dose group, and 24 hours after the last administration. Each group killed 12 animals alive. Test each index. The rest of the animals stopped taking medicine. Continue to observe for 2 weeks. For example, abnormal changes were found in pathological examination 24 h later. The remaining animals were killed alive and the remaining animals were dissected. Focus on the toxic response organ to understand the reversible degree of toxic reaction and possible delayed toxicity.

Results

The rats in the general observation group and the control group had normal activity during the 12 week test period, the behavior was lively, the hair was smooth, the feeding, drinking water and feces were normal. No symptoms of poisoning were found. No death was found in the animals.

Animal body weight growth in 3 groups was basically the same. There was no significant difference between the two groups ($P > 0.05$). Results are shown in Table 1.

TABLE 1. Effects of Kangxuan granules on body weight growth in rats for 12 weeks ($\bar{x}$±s, n=20)

Time	Control group/g		High dose group/g		Low dose group /g	
	♀	♂	♀	♂	♀	♂
Before the experiment	93.2±5.8	94.8±8.1	93.5±5.9	110.9±12.1	92.9±7.0	94.6±7.2
1 week after medication	108.4±9.9	109.5±12.2	108.4±9.2	110.9±12.1	108.6±9.0	111.3±6.9
2 weeks after medication	123.6±6.6	129.2±13.7	124.5±9.3	128.1±11.5	124.7±11.3	131.2±7.7
3 weeks after the medicine	129.4±6.5	143.8±16.2	139.1±9.8	143.3±14.2	138.3±12.1	147.6±10.9
4 weeks after medication	148.0±8.3	157.3±21.2	152.0±8.2	162.5±16.5	152.9±12.6	169.3±16.5
5 weeks after medication	164.5±10.0	174.1±226.3	171.8±11.0	188.2±19.6	169.0±15.9	194.6±19.6
6 weeks after medication	181.7±10.1	190.2±29.5	197.6±11.0	217.4±21.9	187.3±18.5	227.4±25.2
7 weeks after medication	198.2±9.9	225.7±27.2	201.8±10.1	248.3±20.9	212.9±19.6	260.9±28.5
8 weeks after medication	225.5±14.7	262.1±35.8	219.2±12.4	262.9±22.3	223.9±17.1	287.8±23.3
9 weeks after medication	230.9±13.3	283.9±26.9	230.4±18.4	284.0±19.7	231.0±14.6	309.6±31.5
10 weeks after medication	240.0±18.2	300.6±20.9	236.2±21.5	294.6±20.1	238.4±11.2	330.0±28.7
11 weeks after the medicine	245.6±23.8	312.9±24.3	238.5±17.4	314.7±26.4	243.9±9.1	345.7±31.2
12 weeks after medication	253.5±15	328.9±27.2	246.7±17.1	335.3±31.6	253.4±8.5	360.8±31.6

Note: compared with the control group, the high and low dose groups are*P>0.05.*

Blood samples were taken 24 hours after the last administration in each group and control group. Their HB, RBC, WBA and DC. There was no significant difference between the control group and the control group ($P > 0.05$). Results are shown in Table 2.

TABLE 2. Results of blood routine examination of rats after 12 weeks of oral administration of Kangxuan granules ($\bar{x}$±s,n=12)

Group	RBC /$\times 10^{12} \cdot L^{-1}$	Hb /$g \cdot L^{-1}$	PLT /$\times 10^{9} \cdot L^{-1}$	WBC /$\times 10^{9} \cdot L^{-1}$	classify		
					L	M	C
control group	5.0 ± 1.1	134.8 ± 8.1	552.1 ± 155.6	19.4 ± 3.5	83.2 ± 6.8	5.2 ± 1.6	11.7 ± 5.5
High dose group	4.8 ± 1.1	134.8 ± 11.4	578.7 ± 166.8	18.7 ± 4.1	80.8 ± 8.8	5.7 ± 2.0	13.5 ± 7.1
LDG	5.8 ± 1.0	133.8 ± 7.4	496.0 ± 108.9	16.8 ± 5.6	83.6 ± 4.5	52.8 ± 1.8	11.7 ± 3.8

Note: compared with the control group, the high and low dose groups were all. *P>0.05.*

Blood biochemical indexes were measured by liver, kidney function and blood glucose in each group and control group. The results showed that there was no significant difference between the experimental group and the control group ($P > 0.0$). The results are shown in Table 3.

TABLE 3. Effects of anti dizziness Granule on liver, renal function and blood glucose metabolism after 12 weeks of continuous gastric lavage in rats (x±s, n=12)

Group	Control group	High dose group	Low dose group
ALB/ $g \cdot L^{-1}$	36.17 ± 2.7	37.7 ± 2.7	36.8 ± 2.0
TP/ $g \cdot L^{-1}$	75.4 ± 3.8	74.4 ± 5.6	72.9 ± 5.1
GLB/ m $mol \cdot L^{-1}$	4.6 ± 1.1	5.0 ± 1.2	1.0 ± 1.0
± BUN/ m $mol \cdot L^{-1}$	6.1 ± 10.6	6.0 ± 0.5	6.3 ± 0.8
CRE/μ $mol \cdot L^{-1}$	75.7 ± 5.6	78.2 ± 6.4	79.2 ± 6.0
T-CHO/ m $mol \cdot L^{-1}$	1.55 ± 0.22	1.52 ± 0.23	1.51 ± 0.24
ALT/U·L^{-1}	100.8 ± 21.6	96.2 ± 13.9	94.4 ± 31.7
AST/U·L^{-1}	161.0 ± 40.6	153.1 ± 26.6	148.8 ± 38.0
ALP/U·L^{-1}	168.2 ± 51.2	173.8 ± 57.4	179.8 ± 47.9

Note: compared with the control group, the indexes of the high and low dose groups are all. *P>0.05.*

Autopsy and visceral comparison of experimental rats. Organs of rats in each dose group and control group. Its shape, color and texture were observed with the naked eye. The organs were weighed and the specific weight of organs per 100 g body mass was calculated. The results showed that there was no significant difference between the viscera ratio of each administration group and the control group (see table 4).

TABLE 4. Effects of Kangxuan granules on organ specific quality after continuous gastric perfusion for 12 weeks(x±s,n=12)

Group	Control group	High dose group	Low dose group
brain	0.36 ± 0.05	0.04 ± 0.08	0.43 ± 0.09
heart	0.34 ± 0.05	0.31 ± 0.04	0.34 ± 0.04
liver	3.60 ± 0.53	3.73 ± 0.60	3.07 ± 0.31
spleen	0.37 ± 0.09	0.32 ± 0.07	0.28 ± 0.07
lungs	0.58 ± 0.10	0.62 ± 0.12	0.57 ± 0.16
kidney	0.57 ± 0.07	0.61 ± 0.08	0.60 ± 0.13

Note: compared with the control group, the high and low dose groups were all *P>0.05*

After fixed by histopathological examination, paraffin embedded sections were stained with HE and observed under light microscope. The results showed that there was no significant difference between the high dose group and the control group, and no obvious pathological changes were found.

During the convalescence period, the body mass of the experimental rats increased normally and the organs were observed with naked eyes. Because there is no obvious toxicity in this experiment, animals in convalescence period can be exempted from pathological examination.

DISCUSSION

After 12 weeks of continuous perfusion of anti-vertigo granule, compared with the experimental group, the general state, body mass and main organ coefficient of the control group had no significant difference. The main organs were not toxic to the mass, hemogram, liver and kidney function, blood glucose metabolism and the structure of main organs. Therefore, the anti-vertigo granules were 50 times of the clinical dosage. It is safe and non-toxic to rats. It shows that the toxicity of anti-glare granules is very small, and the dosage is safe.

REFERENCES

1. Administration of Drug Administration of the Ministry of Health of the People's Republic of China. *Guidelines for the Research of New drugs in traditional Chinese Medicine* (Beijing: people's Health Publishing House, Beijing, 1993), pp. 205.
2. Administration of Drug Administration of the Ministry of Health of the People's Republic of China. *Compilation of guidelines for preclinical study of new drugs (western medicine)* (Beijing: people's Health Publishing House, Beijing, 1993), pp. 1994.