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PHALLOIDINE AND CARDIOVASCULAR SYSTEM: *IN VITRO* AND *IN VIVO* STUDIES

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ABSTRACT On isolated hearts phalloidine produced negative inotropic and chronotropic effects. In rats, rapid iv 0.75–1.5 mg/kg caused an arterial hypotension

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and a bradycardia. An ip injection caused marked changes in hepatocytes examined by electron microscopy.

KEY WORDS phalloidine; heart; hypotension; bradycardia; liver cytology

Phalloidine, one of the main toxic components of the poisonous fungus *Amanita phalloides*[1], selectively affected the plasma membrane and the cytoskeleton of hepatocytes, causing rapidly a striking increase of actin filaments and a lethal hemorrhagic dystrophy of of liver. Adult animals died within a few hours after poisoning with phalloidine due to hemorrhagic-hypovolemic shock that occurred concomitantly with the development of a severe hemorrhagic dystrophy of liver[2]. The present work reports investigations conducted with phalloidine on cardiovascular apparatus.

METHODS

Male NOS white rats (230–290 g) and guinea pigs (400—500 g) were fasted but with free access to water for 12 h before the experiments.

Rat hearts were isolated on a Langendorff–Spadolini apparatus and both left and right auricles of guinea pigs were isolated on a Basile 7090 Gemini cardiotachograph and bathed in an oxygenated Ringer–Locke solution at 37°C.

For rats anesthetized with ethyl urethane 0.8 g/kg im, arterial blood pressure in common carotid artery and ECG were recorded respectively by a Hellige polygraph and by a Cardioline Epsilon 2 electrocardiograph. Phalloidine 0.3 ml/rat was injected iv into penis dorsal vein in 5 s.

Statistical evaluation was obtained according to Burn *el al*[3].

RESULTS AND CONCLUSION

In the isolated rat hearts, phalloidine concentrations corresponding to or greater than 10 μg/ml induced a moderate and transitory negative inotropic and chronotropic effect not associated with significant modifications of coronary blood flow. Concentrations of 1 μg/ml were inactive. See Table 1.

On the isolated guinea pig auricles, phalloidine produced a very moderate

Table 1. Maximum % variations on isolated rat hearts and guinea pig auricles. N = 5. $\bar{x} \pm SD$

Phalloidine (μg/ml)	Maximum % variations in rat hearts			Guinea pig auricles	
	Inotropism	Chronotropism	Coronary flow	Inotropism	Chronotropism
0.1	+0.80±0.022	−2.00±0.22	+1.07±0.18	+0.90±0.022	+1.00±0.20
1	—	—	—	+1.00±0.04	+0.80±0.16
10	−14±6	−15±5	+0.80+0.022	−11±6	−11±5
100	−19±5	−19±5	+0.60±0.022	−18±6	−19±8

Table 2. Effects of iv phalloidine on arterial blood pressure (mm Hg). $\bar{x} \pm SD$

Rats	Dose (mg/kg)	0 h	0.5 h	1 h	2 h	5 h	7 h
4	0	100±12	101±7	100±5	99±5	94±4	93±5
8	0.75	102±18	94±5	84±10	71±31	60±40	60±40
8	1	110±17	82±22	82±24	58±28	56±23	

Table 3. Effects of iv phalloidine on cardiac frequencies (R-R') in beats/min. $\bar{x} \pm SD$

Rats	Dose (mg/kg)	0 h	0.5 h	1 h	3 h	5 h	7 h	8 h
4	0	462±12	455±20	452±22	435±12	427±8	415±4	410±8
8	0.75	417±48	393±57	362±76	336±96	334±127	405±45	390±4
8	1	427±42	402±37	355±54	252±51	204±42	162±31	115±5
4	1.5	412±8	352±14	330±34	197±12	127±34		

Table 4. Number of rats died after iv phalloidine

Rats dosed	Dose (mg/kg)	Number of rats died at 3 h	5 h	7 h	8 h
8	0.75	0	1	4	4
8	1.0	0	3	3	4
8	1.5	0	4	8	

negative inotropic and chronotropic effect (Table 1).

In rats the rapid iv of phalloidine 0.75–1.5 mg/kg in 5 s caused a slow and progressive arterial hypotension, initially a sinus bradycardia (with increase of PQ and TP intervals, and of QRS and T waves voltage) and successively A-V blockade with a ventricular rhythm were seen (Table 2,3).

The death of rats was due to an extreme hypotension with bradycardia (ventricular rhythm). The latency and intensity of cardiovascular effects and the survival time were dosedependant (Table 4).

Phalloidine ip 0.75–1 mg/kg in anesthetized rats caused marked modifications in liver examined by electron microscopy. In rats killed 3 h after the ip sublethal doses one can see altered microvilli surrounded by a well developed fibrillar reticulum due to prolieration of actin filaments in some hepatocytes delimitating biliary capillaries. In rats which died 3 h after ip phalloidine 0.75 mg/kg there were sinusoids filled with blood and liver cells having various sizes vacuoles which often enveloped erythrocytes and a fibrin reticulum (socalled hemorrhagic dystrophy of liver). The nuclear lesions that are due to amanitines after poisoning with *Amanita phalloides* were lacking. In rats killed 17 h after ip a sublethal dose of phalloidine (0.1 mg/kg), the erythrocytes were enveloped by vacuoles in liver cells with thrombocytes and fibrin precipitates (as a sign of shock state).

In conclusion our experiments proved that the depressing effects in cardiovascular apparatus were less remarkable than those in liver. Now we have programmed some investigations on dogs with the aim to clarify the effects on various vascular districts.

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