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Antiarrhythmic Activity of N-Deacety lappaconitine when Administered Orally

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Abstract. The aim of the study was to study the antiarrhythmic activity of N-deacetylappaconitine when administered orally. The study was conducted on models of arrhythmias caused by intravenous administration of a solution of aconitine, barium chloride, calcium chloride and epinephrine to animals. N-deacetylappaconitine showed a pronounced antiarrhythmic effect, which significantly exceeded the activity of allapinine in aconitine and barium chloride arrhythmias

Keywords: N-deacetylappaconitine, allapinine, arrhythmia, aconitine, barium chloride, calcium chloride, and epinephrine hydrochloride.

Introduction. Cardiovascular diseases remain the leading cause of death in the world. According to available data, 17.9 million people died from CVD in 2016, accounting for 31% of all deaths worldwide. At the same time, almost 85% of these deaths were the results of a heart attack and stroke [1]. Not the last place in the development of these conditions and their consequences is occupied by heart rhythm disorders. One of the leading causes of death from CVD is sudden cardiac death, the share of which in this category of mortality is 25% [2]. Despite the undoubted success in the development of new antiarrhythmic drugs, which has allowed in recent years to significantly expand their arsenal, the drug correction of arrhythmias remains largely unresolved [3]. All this makes the efforts of modern cardiopharmacologists to find and study new medicines that combine, on the one hand, high efficiency, and, on the other, safety of use undoubtedly relevant. Drug therapy for cardiac arrhythmia is based on the use of 28 highly effective antiarrhythmic drugs, including IC-class drugs. Such representatives of this class as propafenone, flecainide, etacizine, as well as the domestic drug Allapinine have been used for many years as effective medicines for both supraventricular and ventricular arrhythmias. Developed in the late 70s at the Institute of Plant Chemistry of the Academy of Sciences of Uzbekistan and approved for clinical use in 1989, Allapinine, despite its high effectiveness, has some limitations in the use of some patients (6-11. 4%) [4] due to side effects in the form of dizziness, headache and eye fixation. In this regard, there is a need to develop a new drug with

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comparable effectiveness, but with fewer side effects. Many antiarrhythmic drugs undergo active metabolism in the body. Studies have found that allapinine undergoes active metabolism in the body with the formation of 6 products. As a result, all the metabolites of allapinine were isolated and developed in the ICPS of the Academy of Sciences of the Republic of Uzbekistan. The main metabolite of allapinine is N-deacetylappaconitine (N-DAL). The aim is to create a drug N-deacetylappaconitine for oral use, which has an antiarrhythmic effect, low toxicity and a large breadth of pharmacological action. The aim of the work is to determine the antiarrhythmic activity of existing antiarrhythmic drugs from metabolites. Some scientists have studied the protonation of N-deacetylappaconitine in methanol solutions [6].

Materials and methods of research. The experiments were carried out on white mongrel mussels and male rats, respectively, with a weight of 18-22 g, 220-250 g contained in standard vivarium conditions with free access to water and food. All procedures with animals were carried out in accordance with the requirements of the international recommendations of the European Convention for the Protection of Vertebrates Used for Experiments or Other Scientific Purposes [5]. Acute toxicity of the compounds was determined by oral and intravenous administration. The antiarrhythmic activity of N-deacetylappaconitine was evaluated in a number of antiarrhythmic models: aconitine, barium chloride, calcium chloride, and epinephrine. Before starting the experiment, an ECG (II standard lead) is recorded in the animals. Then a dose of aconitine is selected, which in all experiments causes a mixed atrioventricular extrasystole. Aconitine was injected into the caudal vein at a dose of 12-15 mcg/kg. Cardiac arrhythmias, as a rule, begin to form from the first to second minute after the end of the administration of aconitine. ECG registration is carried out 3, 5, 10, 15 and 20 minutes after the administration of aconitine. The activity of the studied compound is evaluated by its ability to prevent heart rhythm disturbances caused by aconitine. Calcium chloride arrhythmia was caused in rats by a single administration of the caudal vein at a dose of 250 mg / kg of body weight, and epinephrine arrhythmia by administration of the caudal vein of epinephrine at a dose of 0.2 mg/kg. Cardiac arrhythmias, as a rule, begin to form from the fifth to tenth minute after the end of the administration of calcium chloride and epinephrine. The studied compound is intragastrically administered prophylactically 60 minutes before the introduction of aconitine. The experiment is carried out according to the scheme of control studies.

At the end of the experiments, the average effective dose of the compound under study is calculated — ED50, i.e., the dose that prevents the development of cardiac arrhythmias in 50% of animals. Data processing is carried out using the Litchfield and Wilcoxon method. In addition, the antiarrhythmic index (therapeutic latitude), i.e. the ratio of LD50 to ED50, is calculated. The

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effectiveness of the studied drugs was evaluated in comparison with allapinine, which is considered as a reference (reference) antiarrhythmic drug from commercially available drugs with high activity.

Results and discussion.

1. Resorptive effect and toxicity.

In experiments on mice and rats, it was found that N-DAL is similar to allapinine in the nature of its resorptive action. Unlike the latter, it is less toxic, has a less pronounced depressing and less prolonged effect. The comparative toxicity of N-DAL and allapinine is shown in Table 1.

From the presented data, it follows that in experiments on mice and rats, DAL is inferior to allapinine in toxicity with intravenous administration by 1.23-1.24 times, with intraperitoneal and intragastric administration by 1.58-1.93 times.

Table 1.

Comparative toxicity of N-deacetylappaconitine (N-DAL) and allapinine

Animal	Method of administration	LD 50 mg/kg (P=0.05)		Toxicity to allapinine
		allapinine	N-DAL	
Mouse	Intravenous	6,1 (4,6+8,1)	7,5 (6,8+8,3)	1,23
	Intraperitoneal	15,5 (12,9+18,6)	30,0 (26,8+33,6)	1,93
	oral	52,0 (43,7+61,9)	82.0 (73,9+91,0)	1,58
Rat	Intravenous	5,9 (4,9+7,1)	7,3 (6,6+8,03)	1,24
	oral	57,5 (51,3+64,4)	110 (98,2+123,2)	1,91

2. Study of the antiarrhythmic activity of N-DAL on a model of aconitine arrhythmia

In experiments on a model of aconite arrhythmia in rats, N-DAL showed high protective and relieving antiarrhythmic activity. As can be seen from the data presented in Tab. 2. According to the ability to prevent and stop heart rhythm disorders in rats induced by aconitine, N-DAL is not inferior and even slightly superior to allapinine. In contrast to allapinine, the use of N-DAL on

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the background of aconite dysarrhythmia leads to a faster relief and restoration of the normal sinus rhythm.

So, if the maximum effect after oral administration of N-DAL at a dose of 0.05-0.1 mg/kg occurs in 30 minutes, reaches a maximum in 50-60 minutes and lasts 8 hours or more, then the restoration of sinus rhythm in animals treated with allapinine (0.1 mg/kg) when ingested, the effect develops in 40-60 minutes, reaches a maximum in 80 minutes and lasts 8 hours or more.

Table 2.

Antiarrhythmic efficacy of N-deacetylappaconitine (N-DAL) at different doses in aconitine-induced arrhythmia in rats (n=10).

№	Name of the drug	Doses of mg / kg	Violation of the heart rhythm			Survival rate %	
			Arrhythmia %	Start in min.	Duration in min.	survived	fall down
1.	Control + Aconitine	15 mkg/kg	100	2,83	Not recovered	20	80
2.	N-DAL + Aconitine	0,01	100	4,8	Not recovered	20	80
		0,05	80	6,4	18	60	40
		0,1	50	9,3	9	80	20
		0,5	20	11,2	6	100	0
		1,0	0	-	-	100	0
3.	Allapinine + Aconitine	0,05	100	2,1	Not recovered	0	100
		0,1	80	4,7	Not recovered	20	80
		0,5	70	8,4	15	50	50
		1,0	50	9,7	11	70	30
		1,5	0	-	-	100	0

A comparison of antiarrhythmic index (LD50/ED50) compare drugs suggests that N-DAL has an advantage over allapinine and the breadth of therapeutic action (table 3).

From the data of table 3. it follows that the antiarrhythmic index N-DAL with prophylactic use of inside is equal to 1183, the same indicators for allapinine are respectively 115. N-DAL issued when the application inside superior allapinine as 10 times the oral and parenteral administration of more than 1.5 times.

Consequently, in experiments on the model of aconite arrhythmia in rats, N-DAL has a powerful antiarrhythmic effect. In terms of antiarrhythmic activity, N-DAL is not inferior to allapinine and the known antiarrhythmic agents are superior to it in terms of the speed of the

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antiarrhythmic effect and the breadth of the therapeutic effect.

Table 3.

Comparative antiarrhythmic efficacy N-deacetylappaconitine (N–DAL) and allapinine in aconitine-induced arrhythmia in rats

Preparation and method of use	LD50, mg/kg inside	ED50, mg / kg when used	Antiarrhythmic index (LD50/ED50) when used
		preventive maintenance	preventive maintenance
N-DAL (inside)	110 (98,2±123,2)	0,093 (0,04±0,12)	1183
Allapinine (inside)	57,5 (51,3±64,4)	0,5 (0,12±0,92)	115

Table 4.

Comparative antiarrhythmic efficacy of N-deacetylappaconitine (N-DAL) and known antiarrhythmic agents in a rat model of aconitine arrhythmia.

Preparation	Inside		
	ED 50 mg / kg	LD50 mg / kg	Antiarrhythmic index LD50/ ED50
N-DAL	0,093	110,0	1183
Allapinine	0,5	57,5	115,0
Etmozin	57,5	1025,0	17,0

The high antiarrhythmic activity of N-DAL, compared with the activity of allapinine, is also shown in other experimental models of heart rhythm pathology.

3. Study of the antiarrhythmic activity of N-DAL on a barium chloride model of heart rhythm disorders

It is known that barium chloride is able to inhibit potassium conductivity. In accordance with this, the model of arrhythmia using barium chloride is adequate to identify substances with class III antiarrhythmic properties.

Table 5.

Antiarrhythmic efficacy of N-deacetylappaconitine (N-DAL) at different doses in barium chloride-induced arrhythmia in rats (n=10)

№	Name of the drug	Doses of	Violation of the heart rhythm	Survival rate %

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		mg / kg	Arrhythmia %	Start in sec.	Duration in min.	survived	fall down
1.	Control Barium Chloride	15 mg / kg	100	5,83	12	50	50
2.	N-DAL + Barium Chloride	0,01	100	9,2	12	50	50
		0,025	100	11,6	9,2	60	40
		0,05	80	17	6,75	80	20
		0,1	60	21	4,23	100	0
		0,5	50	23,75	2,75	100	0
		1,0	40	20	1,25	100	0
3.	Allapinine + barium chloride	0,1	100	20,2	12	50	50
		0,5	60	24,3	11,7	40	60
		0,75	40	26	10,8	60	40
		1,0	0	0	0	100	0

So, N is GIVEN in doses from 0.01 to 1 mg/kg (orally) warns in 50-100% of rats ($P < 0.001$) in the occurrence of ventricular arrhythmia induced by the introduction of toxic doses of barium chloride (15 mg/kg), at doses of 0.2-0.5 mg/kg completely eliminated from anesthetized cats and dogs fibrillation and hypotension caused by electrical irritation about threshold currents of ear right atrium and (or) the apex of the left ventricle, in doses of 0.05-0.1 mg/kg inhibits 80-100% ventricular.

Table 5 shows that N-DAL in the oral administration of barium chloride-induced arrhythmia is somewhat not inferior to allapinine in antiarrhythmic activity, so it has a higher antiarrhythmic index for preventive use (Table 6).

Table 6.

Comparative antiarrhythmic efficacy N-deacetylappaconitine (DAL) and allapinine in barium chloride-induced arrhythmia in rats

Preparation and method of use	LD50, mg/kg inside	ED50, mg / kg when used	Antiarrhythmic index (LD50/ED50) when used
		preventive maintenance	preventive maintenance
N-DAL (inside)	110 (98,2±123,2)	0,033 (0,013±0,067)	3333,33

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Allapinine (inside)	57,5 (51,3±64,4)	0,805 (0,706±0,917)	71,4

Study of the antiarrhythmic activity of N-DAL in a dose 0,05-0,1-0,5-1,0 mg / kg on calcium chloride and epinephrine models of cardiac arrhythmias in anesthetized rats did not eliminate arrhythmia and survival.

Conclusion. N-DAL in antiarrhythmic activity is not inferior to allapinine in contrast to it, it is 2 times less toxic, has a greater therapeutic breadth and, most importantly, 3 times has a faster stopping antiarrhythmic effect, which allows it to be used in preventive and urgent situations.

Thus, the results of the study allowed us to establish that the studied substances have antiarrhythmic activity in the conditions of aconitine and barium chloride models of arrhythmia. As is known, the mechanism of occurrence of arrhythmia under the influence of aconitine is associated with a change in the properties of fast sodium channels of the excitable membrane and an increase in their conductivity.

It was also found that N-DAL has a pronounced antiarrhythmic activity and has an advantage in the breadth of therapeutic action over the known class I antiarrhythmic drugs (allapinin, etmosin).

It is known that barium chloride is able to inhibit potassium conductivity. In accordance with this, the model of arrhythmia using barium chloride is adequate to identify substances with class III antiarrhythmic properties. N-DAL in the oral management of barium chloride-induced arrhythmia in antiarrhythmic activity is somewhat not inferior to allapinine, so it has a higher antiarrhythmic index of preventive use.

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