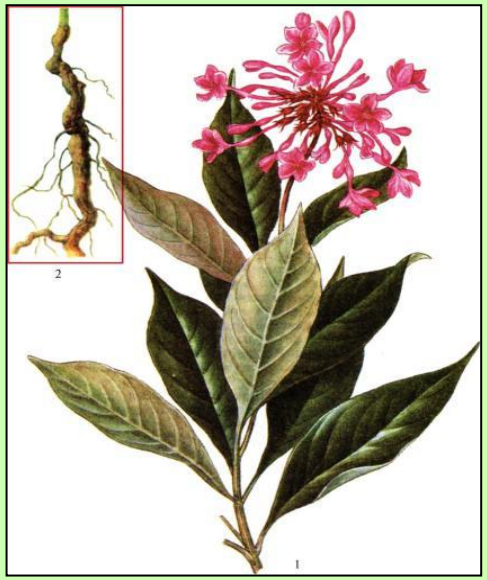
PRODUCTIVITY, ANTIARRHYTHMIC AND HYPOTENSIVE ACTIVITY OF ALKALOIDS FROM *RAUVOLFIA SERPENTINA* TISSUE CULTURE

Konvalyuk I.I.[1], Dobrelia N.V.[2], Khromov O.S.[2], Soloviev A.I.[2], Bieda O.A.[1,3], Mozhylevska L.P.[1], Yarmoluk S.M.[1], Kunakh V.A.[1]

[1]*Institute of Molecular Biology and Genetics (IMBG) of the NAS of Ukraine*
[2]*State Institution "Institute of Pharmacology and Toxicology of the NAMS of Ukraine“,*
[3]*Scientific service company “LLC OTAVA”, Ukraine*

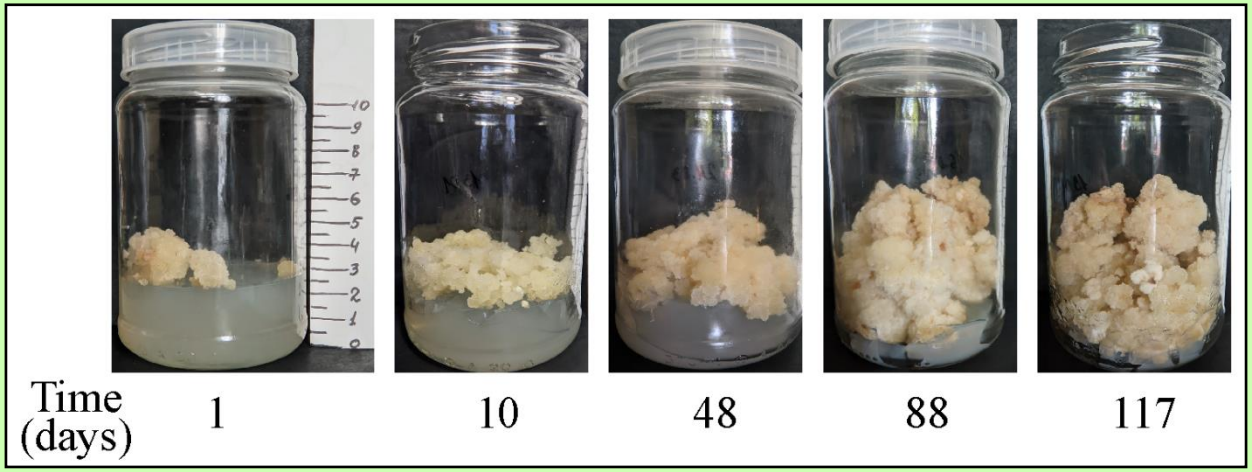


Rauvolfia serpentina

Rauvolfia serpentina is a medicinal tropical plant widely used in traditional medicine of India and China due to its antiarrhythmic, hypotensive, sedative, and psychotropic activities. Considering the scarcity of natural resources, the strain K-27M of *R. serpentina* tissue culture has been established at the IMBG of the NAS of Ukraine.

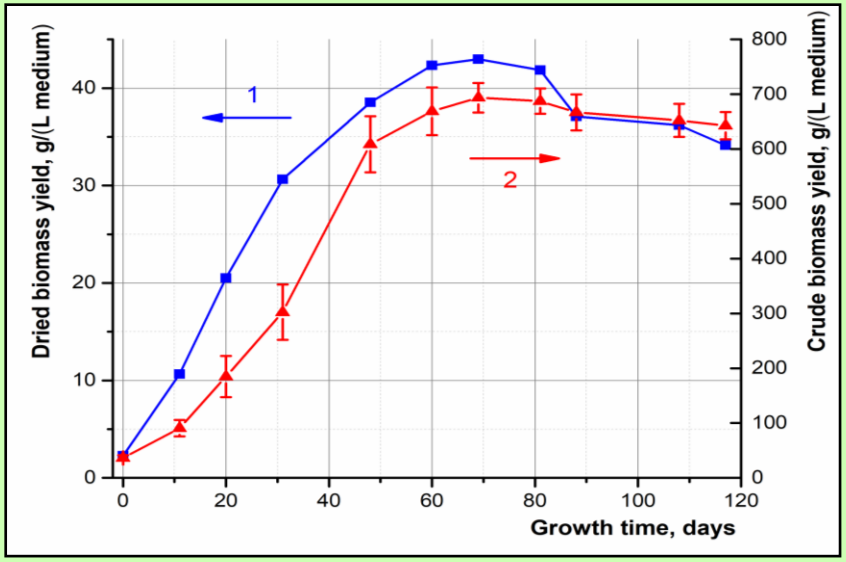
The ***aim*** was to investigate the productivity of the strain and the content of indole alkaloids, as well as to evaluate the antiarrhythmic and hypotensive effects of various fractions of biomass extracts of the *R. serpentina* K-27M strain.

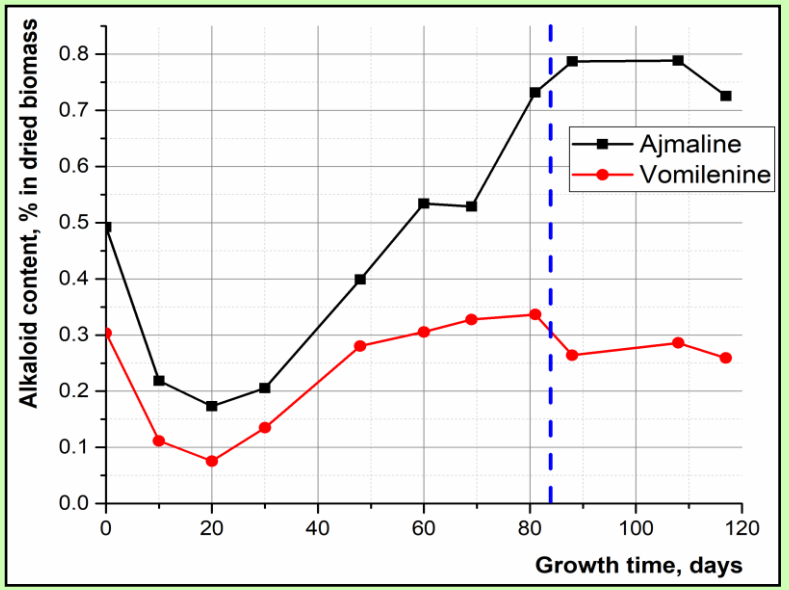
Methods. The strain was cultured at 27–28°C in the dark on the 10C solid agar medium without growth regulators. The content of indole alkaloids was determined using HPLC-MS.

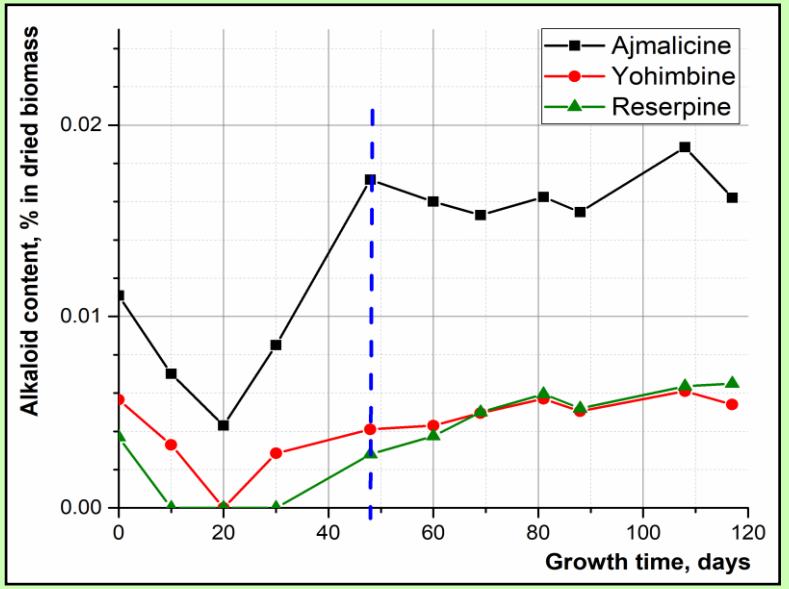


General appearance of the K-27M strain of *R. serpentina* tissue culture grown on the 10C medium at different time points after subculture.

The hypotensive activity was evaluated using the rat thoracic smooth muscle *in vitro* and by assessing the effect of intravenous administration on blood pressure *in vivo*. The antiarrhythmic activity was studied in rats with models of adrenaline-induced arrhythmias as well as on the isolated guinea pig hearts with ischemia and reperfusion-induced arrhythmias.







Results. 1) The maximum wet biomass yield was at 69th day of subculture and amounted to 670-690g/L medium of wet biomass or 40-43g/L of dry biomass.

2) The dry biomass contained 4.0% of total indole alkaloids, including 1.64% of ajmaline-like alkaloids, 0.79% of ajmaline, 0.34% of vomilenine, and 0.006% each of yohimbine and reserpine.

Cell biomass of the K-27M strain

Dry biomass

Fraction 1
Extraction with 25% ethanol.
Content: total alkaloids 2.2%: ajmaline 1.5%, acetyl-ajmaline 55%, raucafricine 10%.

Fraction 2
Extraction with 70% ethanol.
Content: total alkaloids 6.4%; mainly ajmaline 2.2%, raucafricine 1.5%, acetyl-ajmaline 1.4%.

Fraction 3
Extraction with ethyl acetate.
Content: total alkaloids 29.0%; mainly ajmaline 11%, vomilenine 5.1%, acetyl-ajmaline 5%.

Wet biomass

Fraction 4
Content: total alkaloids 65.0%; mainly vomilenine 42.5%*, acetyl-ajmaline 9.1%*.

Fraction 5
Content: total alkaloids 47.4%; mainly acetyl-ajmaline 33.0%, vomilenine 6.4%.

3) Five fractions were obtained from the extracts of dry (fractions 1,2,3) and wet biomass (fractions 4,5) of K-27M strain.

Effect of intravenous administration of ajmaline and fractions of *R. serpentina* alkaloids on changes in the QTc interval (ms) in rats

Group		Initial value	30 min
Ajmaline (n = 6)	m ± SE	131,44 ± 12,77	41,67 ± 4,30
	p		< 0,01
Fraction 1 (n = 6)	m ± SE	139,20 ± 3,89	160,58 ± 2,94
	p		> 0,05
	p1	> 0,05	< 0,001
Fraction 2 (n = 6)	m ± SE	139,07 ± 9,38	149,10 ± 5,36
	p		> 0,05
	p1	> 0,05	< 0,001
Fraction 3 (n = 6)	m ± SE	138,99 ± 8,19	121,62 ± 4,46
	p		> 0,05
	p1	> 0,05	< 0,001
Fraction 4 (n = 6)	m ± SE	113,06 ± 11,14	104,55 ± 7,07
	p		< 0,05
	p1	< 0,01	< 0,05
Fraction 5 (n = 6)	m ± SE	125,88 ±10,36	92,49 ± 10,73
	p		< 0,05
	p1	> 0,05	< 0,05
	p2	> 0,05	< 0,05

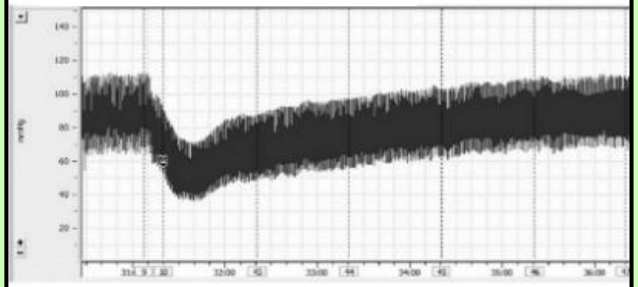
Notes: p – compared to the initial value; p1 - compared to ajmaline; p2 - compared to fraction 4.

4) The fractions 4 and 5 had a pronounced antiarrhythmic effect in rats. In contrast, fractions 1 and 2 depleted of indole alkaloid showed a weak proarrhythmic effect. The antiarrhythmic effect of fraction 5 on the isolated guinea pig hearts was comparable to the control sample of ajmaline in terms of the activity intensity.

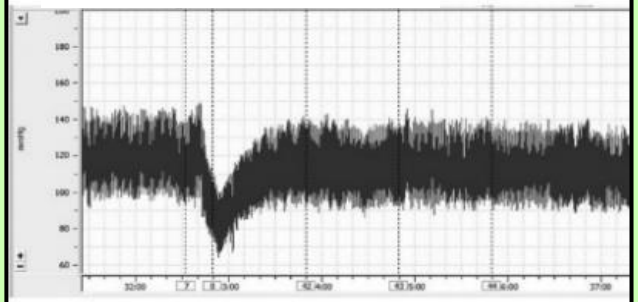
5) Fractions 1 and 2 exerted a constrictor effect on the rings of the thoracic aorta of rats. The fractions 4 and 5 had a vasorelaxing effect *in vitro* and transitory (15-30s) hypotensive effect *in vivo*.

Blood pressure changes under the influence of ajmaline and fractions 4 and 5

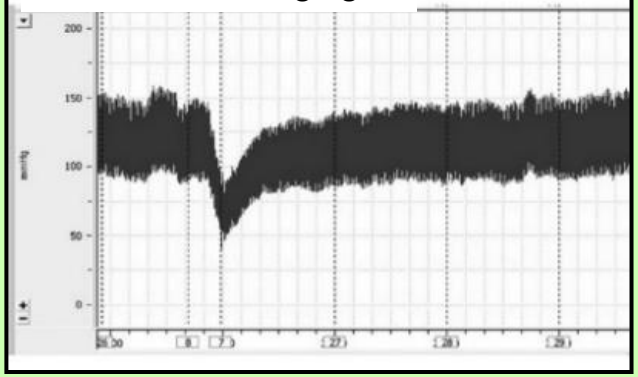
Ajmaline, 2,8 mg/kg



Fraction 4, 2,8 mg/kg



Fraction 5, 2,8 mg/kg



Conclusions. Thus, the studied K-27M strain of *R.serpentina* tissue culture is a promising producer of indole alkaloids. The physiological mode of action depended on the content and composition of alkaloids in the fractions. The wet biomass extracts have demonstrated significant antiarrhythmic and transitory hypotensive effects.