Effect of Pectin Substances on Contractile Activity of the Uterine Myometrium in Rats

A. B. Vyshtakalyuk, N. A. Sosnina, S. T. Minzanova, V. V. Zobov, A. V. Lantsova, E. R. Minullina, V. F. Mironov, and A. N. Karaseva

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 141, No. 4, pp. 414-417, April, 2006
Original article submitted July 20, 2004

We studied the effects of peptides from *Amaranthus cruentus L.*, *Rauwolfia serpentina Benth.*, and citrus plants on contractile activity of the uterine myometrium in rats. Pectin substances inhibited spontaneous contractile activity of uterine muscles and abolished the stimulatory effect of oxytocin. Pectins had a synergistic effect with epinephrine and acetylcholine.

Key Words: *pectin; contractile activity; smooth muscles; uterus; amaranth*

Pectin substances from plants have a wide range of biological properties, which is related to their unique physicochemical activity [6,8,9]. Studying of the effect of pectins on muscle tissue will expand our knowledge of the mechanism of their action. Experiments on isolated rat hearts [2] showed that amaranth pectin causes coronary vasospasm by affecting the tone of smooth muscles in the vascular wall, but has no effect on myocardial contraction.

Published data show that pectin substances from various raw materials differ by the molecular weight, esterification, and structure of carbohydrate side chains [3,7]. These specific features determine differences in physicochemical properties and biological activity of pectins [4]. It is important to evaluate the relationships between physicochemical properties and biological activity of pectin substances from various raw materials.

Here we studied the effect of pectins from various raw materials on contractile activity of smooth muscles in rat uterus.

A. E. Arbuzov Institute of Organic and Physical Chemistry, Kazan Research Center, Russian Academy of Sciences. ***Address for correspondence:*** vysh@iops.kcn.ru. A. B. Vyshtakalyuk

MATERIALS AND METHODS

Experiments were performed with pectins from amaranth plants (*Amaranthus cruentus L.*, AP) and Indian snake root (*Rauwolfia serpentina Benth.*, RSP) and commercial citrus pectin (Classic C-401 pectin, Herbstreith & Fox Company, CP). Characteristics of pectins according to GOST 29186 are presented in Table 1.

The effects of pectin substances on spontaneous contractile activity of the uterine myometrium in rats were studied as described elsewhere [5]. Experiments were performed on 80 intact female outbred albino rats weighing 180-250 g. The animals were decapitated. Each uterine horn was isolated and maintained in a bath with de Gaulon solution at 37°C under constant aeration (95% O_2 and 5% CO_2, pH 7.2-7.4). The prepared uterine horn was mounted on hooks so that one end of the preparation was firmly fixed, while the other end was attached to an isotonic transducer with a ligature. The preparation was stretched to a desired tension (initial load 1 g) for 40-60 min. Contractions of the muscle preparation were recorded with an isotonic transducer and an Ugo Basile device (tape speed 5 mm/min).

The test substances (0.1% solution of pectin substances) were added to a bath with 1 ml pre-

0007-4888/06/1414-0430

TABLE 1. Main Characteristics of Test Pectins

Parameter	AP	CP	RSP
Free carboxylic groups, %	4.5	3.5	0.9
Molecular weight, arb. units (viscosimetry)	85,000-90,000	17,600	100,000-105,000
Uronide constituent, %	70.0-76.0	60.0	96.6
Etherification, %	70.0-75.0	70.0-75.0	50.0

paration. Variations in contractile activity of the uterine myometrium were recorded. After treatment with the test substances, the frequency and amplitude of contractions of rat uterine horn were expressed in percents of baseline spontaneous contractile activity of uterine muscles. We studied the effect of pectins on activity of agonists (epinephrine, acetylcholine, and oxytocin). Agonists in the effective doses were added 10 min after pectin treatment. Otherwise, we administered a mixture of pectins and agonists.

The results were analyzed by Student's *t* test (Origin 6.0 software).

RESULTS

Experiments with active myometrium showed that the test pectins decreases spontaneous contractile activity of the smooth muscle preparation. We observed a decrease in the frequency and amplitude of contractions. CP was most potent in this respect. This pectin decreased the amplitude and frequency of contractions by 26.4±10.3 and 49.2±10.9%, respectively. Variations in the amplitude and frequency of contractions were least significant after treatment with AP and RSP, respectively.

Oxytocin in a dose of 3×10^{-5} g/ml increased the amplitude and frequency of contractions of the uterine myometrium by 53.4±3.3 and 65.0±4.4%, respectively. Long-term application of oxytocin induced spastic contractions of the muscle. Combined treatment with pectins and oxytocin decreased the stimulatory effect of oxytocin on the frequency and amplitude of contractions ($p<0.001$, Fig. 1). RSP was most potent in this respect. This pectin decreased the amplitude and frequency of contractions by 49.5±2.3 and 71.9±1.4%, respectively (compared to individual treatment with oxytocin).

Acetylcholine in a concentration of 1.3×10^{-6} g/ml produced a spasmogenic effect and sometimes caused muscle contracture. Contractile activity was preserved under these conditions. The frequency of contractions increased more significantly than the amplitude (by 44.6±7.2 and 11±3%, respectively). Pectins potentiated the effects of acetylcholine, which manifested in an increase in contraction amplitude (AP, $p<0.05$; RSP, $p<0.01$). The frequency of contractions remained unchanged under these conditions (Fig. 2).

Epinephrine in a concentration of 3×10^{-9} g/ml inhibited spontaneous contractile activity of the isolated uterine horn. Rhythmic activity of smooth muscles recovered 3-4 min after epinephrine administration. However, the amplitude and frequency of contractions remained below the baseline level (by 88.8±0.3 and 60.4±1.7%, respectively). The time-to-recovery of contractile activity was 2-fold longer in experiments with administration of epinephrine after application of pectin substances (6-7 min). The amplitude and frequency of contractions decreased more significantly after combined treatment with epinephrine and pectins (compared to individual administration of epinephrine). Contractile activity of the uterine myometrium was sometimes completely suppressed under these conditions. Differences were revealed in the effect of the test pectins. CP had the most significant effect and decreased the amplitude and frequency of contractions by 98.8±0.3 and 83.4±0.3%, respectively (Fig. 3). The mixture of CP and epinephrine had a more significant effect compared to that observed

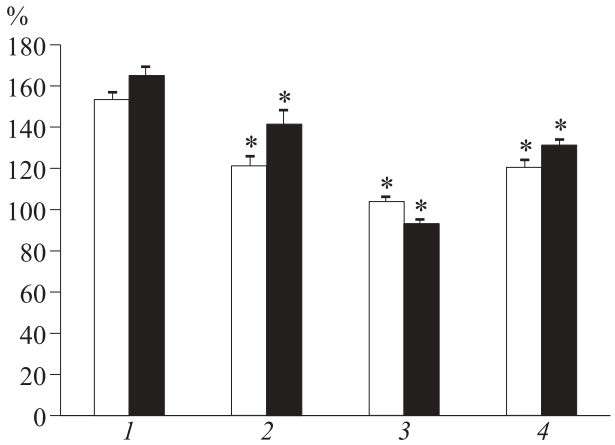

Fig. 1. Amplitude (light bars) and frequency (dark bars) of spontaneous contractile activity of the uterine myometrium under the influence of oxytocin (Ox) and various pectins. Control (Ox, *1*); amaranth pectin (AP, *2*); *Rauwolfia serpentine* pectin (RSP) and Ox (*3*); citrus pectin (CP) and Ox (*4*). *$p<0.001$ compared to the control. Here and in Figs. 2 and 3: baseline activity of the smooth muscle preparation is taken as 100%.

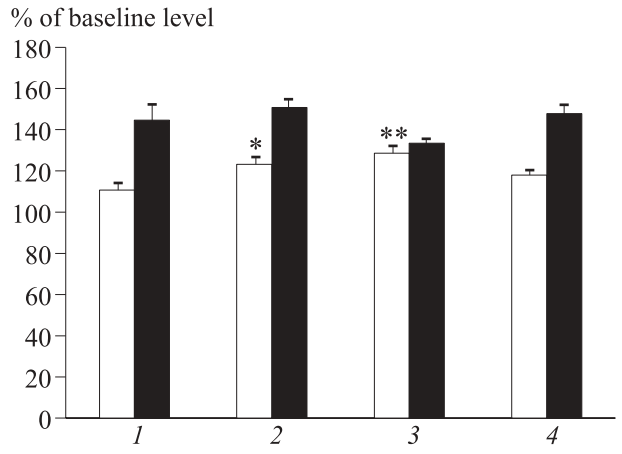

Fig. 2. Amplitude (light bars) and frequency (dark bars) of spontaneous contractile activity of the uterine myometrium under the influence of acetylcholine (ACh) and various pectins. Control (ACh, *1*); AP+ACh (*2*); RSP+ACh (*3*); CP+ACh (*4*). **p*<0.05 and ***p*<0.01 compared to the control.

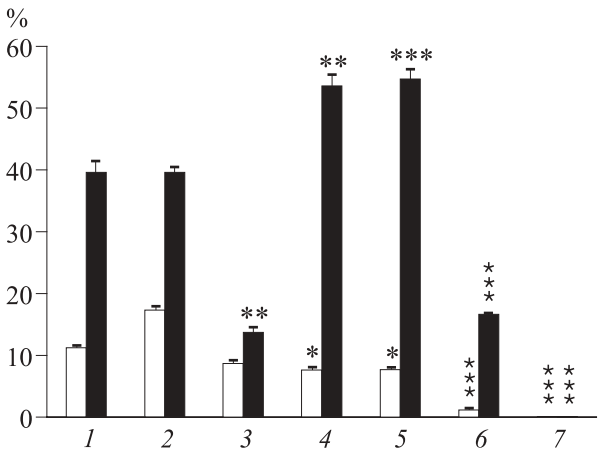

Fig. 3. Amplitude (light bars) and frequency (dark bars) of spontaneous contractile activity of the uterine myometrium under the influence of epinephrine (E) and various pectins. Control (E, *1*); AP (*2*); AP+E (after preapplication, *3*); RSP (*4*); RSP+E (after preapplication, *5*); CP (*6*); CP+E (after preapplication, *7*). **p*<0.05, ***p*<0.01, and ****p*<0.001 compared to the control.

under the influence of epinephrine after preapplication of pectin. This mixture caused relaxation of uterine muscles (Fig. 3).

These data expand our knowledge of physiological activity of plant pectin polysaccharides. We studied the effect of pectins on contractile activity of uterine smooth muscles and evaluated the sensitivity of receptors in the isolated rat uterus to agonists. Pectins possessed activity relative to receptors in uterine smooth muscles. Previous studies demonstrated that pectins have a spasmogenic effect on smooth muscles in the vascular wall [2]. Our experiments showed that pectins inhibit contractile activity of smooth muscle preparations from the uterus. Pectins had a synergistic effect with epinephrine. It can be hypothesized that the effect of pectin substances on smooth muscles is mediated via adrenoceptors. Differences in the influence of pectins on uterine muscles and vascular wall can be related to the prevalence of β_2-adrenoceptors in the uterine myometrium of intact rats. Excitation of these receptors is followed by muscle relaxation [1,5]. By contrast, the vascular wall mainly includes excitation α-adrenoceptors.

It should be emphasized that the effect of pectins correlates with their molecular weight. CP with minimum molecular weight exhibits strong inhibitory activity and had a synergistic effect with epinephrine. This correlation between the effect and molecular weight is consistent with published data [4]. RSP is characterized by a small number of free carboxylic groups, low degree of etherification, and high content of uronide. This pectin is antagonistic to oxytocin and has the lowest synergistic effect with epinephrine.

This work was supported by the Russian Foundation for Basic Research (grant No. 03-03-96224).

REFERENCES

1. O. M. Avakyan, *Sympathoadrenal System* [in Russian], Leningrad (1977).
2. T. L. Desalen', O. V. Tsepaeva, N. A. Sosnina, *et al*., *Byull. Eksp. Biol. Med*., **123**, No. 1, 90-95 (1997).
3. V. I. Maksimov, V. M. Bondarenko, and V. E. Rodoman, *Zh. Mikrobiol. Epidemiol. Immunol*., No. 6, 107-108 (1998).
4. Yu. S. Ovodov, *Bioorgan. Khim*., **24**, No. 7, 483-501 (1998).
5. R. Blattner, H. Classen, H. Denert, and H. Doering, *Experiments on Isolated Preparations of Smooth Muscles* [in Russian], Moscow (1986).
6. J. Duan, X. Wang, Q. Dong, *et al*., *Carbohydr. Res*., **338**, No. 12, 1291-1297 (2003).
7. B. L. Ridley, M. A. O'Neill, and D. Mohnen, *Phytochemistry*, **57**, No. 6, 929-967 (2001).
8. A. H. Terpstra, J. A. Lapre, H. T. de Vries, and A. C. Beynen, *Eur. J. Nutr*., **41**, No. 1, 19-26 (2002).
9. X. S. Wang, Q. Dong, J. P. Zuo, and J. N. Fang, *Carbohydr. Res*., **338**, No. 22, 2393-2402 (2003).