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In vivo and in vitro study of the influence of the anticholinesterase drug galantamine on motor and evacuative functions of rat gastrointestinal tract

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Abstract

Galantamine is efficacious for vascular dementia and Alzheimer's disease. Its application leads to some negative gastrointestinal side effects. The present study observes galantamine-induced influence on gastrointestinal motility of rats and its effects on isolated gastrointestinal smooth muscles. The gastrointestinal tract was studied by X-ray contrast examination. Functional disturbances were observed: hypertonia, increased stomach and ileal peristalsis activity, accelerated intestinal passage. In vitro, the drug caused tonic contractions in smooth muscle preparations and increased the gastric and ileal phasic amplitude. The jejunal smooth muscle strips demonstrated an opposite tendency. The reactions were a result of the interaction of galantamine-accumulated endogenic acetylcholine with M- and N-acetylcholine receptors. The tonic effects were influenced in varying degree by atropine and ipratropium, whereas the phasic by atropine, ipratropium, hexametonium and methysergide. In conclusion, the in vitro effects registered satisfactorily explain in vivo examined galantamine-induced changes in the gastrointestinal tract of the treated rats and can be considered as main cause for development of such changes.

Keywords: Galantamine; Gastrointestinal tract; Acetylcholine; Acetylcholine receptor

1. Introduction

Galantamine is a tertiary alkaloid and a reversible, competitive acetylcholinesterase inhibitor (Zarotsky et al., 2001). The drug is effective and well tolerated for symptomatic treatment of Alzheimer's disease and other forms of dementia which improves cognition, function and daily life activities of patients (Scott and Goa, 2000; Corey-Bloom, 2003). The decrease of acetylcholine deficit in the brain of patients is à basic effect of the drug in these diseases.

Galantamine possesses some other effects as well. It interacts allosterically with nicotinic (N) acetylcholine receptors to potentiate the action of agonists at these receptors (Maelicke, 2000). The drug is a selective inhibitor for acetylcholinesterase rather than butyrylcholinesterase (Darvesh et al., 2003).

Galantamine influences a number of other tissues, organs and systems outside the central nervous system (CNS) in Alzheimer's disease treatment. This is a cause for the unfavorable side effects. Peripheral adverse effects are characteristic of galantamine with an incidence ranging between 7% and 30%. They occur comparatively often in the gastrointestinal tract as abdominal pains (Nordberg and Svensson, 1999), nausea and vomiting (Sramek et al., 2000; Poirier, 2002; Fulton and Benfield, 1996), diarrhea (Nordberg and Svensson, 1999; Cummings, 2003). These effects are connected to a significant degree with the motor function of the stomach and intestines. Their appearance is a consequence of the drug influence on gastrointestinal motility.

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The purpose of the present study was to determine the character and causes for galantamine effects on gastric and intestinal smooth muscle contractile activity, as well as on changes in evacuator and motor functions of gastrointestinal tract of galantamine treated rats.

2. Material and methods

2.1. X-ray contrast examination

Twenty-five male Wistar white rats (group I with 12 control animals, group II with 13 animals, treated with galantamine, weight 310±20 g) were examined. For the period of the experiment (20 days), they were kept at standard life conditions: diet, temperature, dark/light-regime. According to the European Animal Guide Standards, the animals were food deprived for 24 h before the onset of the experiment. The rats from 2nd group received galantamine (a single dose of 1 mg/kg per os) for 20 days. The control rats (group I) were treated with an identical amount of saline only.

The plain facial radiographs of the gastrointestinal tract were performed using a Progress 800ST (CGR) X-ray apparatus with a constant focus distance of 0.7 m. The contrast medium (barium sulphate—120 mg dissolved in 200 ml H_2O, t°=37 °C) was probed per os in a dose of 2.5 ml per animal. The radiographs were taken immediately and at the 1st, 2nd, 3rd, 6th, 12th, 18th, 24th, 26th and 28th hour following introduction of the contrast medium. The time for the complete evacuation of the barium enema out of the stomach and the intestinal passage at the 24th hour were used as radiologically perceptible quantitative indices. The differences in the gastric and intestinal shape, size, tone and peristaltic activity in the two animal groups examined were also recorded.

2.2. Measurement of the mechanical smooth muscle activity

Circular smooth muscle preparations (1×15 mm) from gastric corpus and longitudinal ones from duodenum, jejunum and ileum were used. The smooth muscle strips were fixed at one end on a glass holder. The other end was connected with catgut to Swema tensotransducers (Sweden). In such a way the mechanical activity of the preparations was registered. During the experiment, the smooth muscle strips were flushed out with a Krebs solution bubbled with O_2/CO_2 (19/1 v/v) and maintained at 36 °C.

The contractile activity was recorded with a Linseis recorder (Germany). The tonic contractions were registered as a relative tonic change of smooth muscle preparations before and after each drug application. The changes of the phasic activity were determined as a difference between the mean values of the amplitude of 15 consequent phasic contractions immediately before the application (controls) and after reaching maximum effect after the agent action.

2.3. Drugs, solutions, chemicals

The following chemical substances and drugs were used in the experiment: Galantamine (Nivalin, Sopharma), Ipratropium bromide, Methysergide and Hexametonium chloride (Sigma), Acetylcholine and Atropine (Sopharma), Serotonin (5-hydroxitriptamin, Biotika). The contrast medium was Barium sulphate (Sopharma).

All substances in the Krebs solution were Merck's. The Krebs used in our in vitro experiments had the following composition in mM: NaCl—120; KCl—5,9; $CaCl_2$—2.5; Mg Cl_2—1.2; NaH_2PO_4—1,2; $NaHCO_3$—15.4 and glucose—11.5, pH 7.23–7.26. The pH of the solutions was measured by Microcomputer pH-meter 6201 (Jenco Electronics).

2.4. Statistical analysis

The values obtained were expressed as mean±S.E.M. The comparison between the groups was made by Student's *t*-test of analysis of variance (ANOVA), in the INSTAT computer program. A value of $P<0.05$ was considered as a significant difference.

3. Results

3.1. Galantamine-induced effects on gastrointestinal motility and evacuative activity

The stomachs and the small intestines of control animals showed normal sizes and shapes. Their anatomic parts were visibly distinguished. The peristaltic waves were deep and symmetrical. Immediately following its application in control rats, the contrast medium filled up

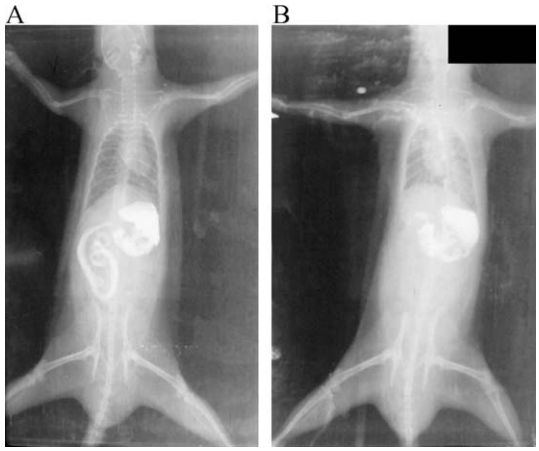

Fig. 1. Contrast facial radiographs of control rat (A) and galantamine-treated rat (B), made immediately after a probe of 2.5 ml $BaSO_4$.

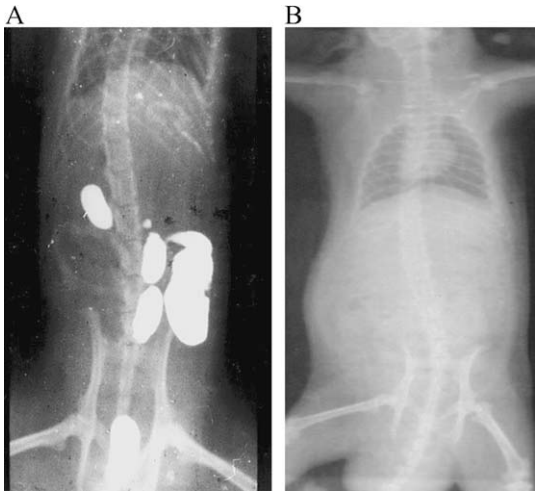

Fig. 2. Contrast facial radiographs of control rat (A) and galantamine-treated rat (B), made on the 24th hour after a probe of enema.

the stomach, duodenum and the initial part of the jejunum (Fig. 1A). An hour later, it was fully evacuated from the stomach of 10 of the controls and distributed along the small intestines (in two stomachs only there were traces of contrast medium). The small intestines had common location and width of the lumen. At the 3rd hour following contrast, the $BaSO_4$ left the stomachs of all control animals and reached the caecum of one of them. At the 6th hour, the enema was evacuated from the small intestines and began to fill the caecum. At the 12th hour, the contrast matter was distributed in the caecum and rectum. At the 18th hour, it was evacuated from the caecum and filled the rats' rectum. At the 24th hour, the barium suspension was fully evacuated from the gastrointestinal tract of four animals. In the others, there were traces in the rectum.

Significant radiological differences were observed in galantamine-treated animals. The stomachs had ordinary shape, location, sharp outlines and distinguishable antral and fornical parts. The radiographs demonstrated some variety in the distribution of the contrast medium in the gastrointestinal tract, immediately after probing: it filled up the stomachs of nine animals (Fig. 1B), reached the duodenal arc of three and filled up the duodenum in one animal. Along the duodenum and the jejunum in some animals, there were narrowed lumen areas, which were preserved on the X-rays from the 3rd hour as well. At the 3rd hour following contrast, the latter was evacuated from the stomachs of a part of the rats. In the others, it was distributed in the stomach and throughout the small intestines. Very well expressed peristaltic activity was observed in the ileal zone. At the 6th hour, the contrast medium was evacuated from all rats' stomachs and from the small intestines of most of rats. It filled the caecum, and in four rats it was observed in the rectum. On the X-rays taken at the 12th hour, the contrast was registered in the rectum. At the 18th hour, it was fully evacuated from the gastrointestinal tract of six rats. At the 24th hour, no traces of the contrast medium were discovered. Most of the galantamine-treated rats had diarrhea on the experimental day, except four of them. A typical intestinal passage at the 24th hour of the two groups of rats was shown (Fig. 2A,B). The quantitative radiological differences characterizing the gastrointestinal function of both experimental groups are presented in Table 1.

Table 1
Time parameters, reflecting the evacuation kinetics of contrast media ($BaSO_4$) in rat gastrointestinal tract

Index	Control group (n=12)	Galantamine-treated group (n=13)
Complete stomach evacuation, h	1.33±0.46	3.46±1.19[a]
Complete gastrointestinal tract evacuation, h	25.67±1.38	20.54±2.21[a]

The comparison is made between control and galantamine-treated group.
[a] $P<0.05$.

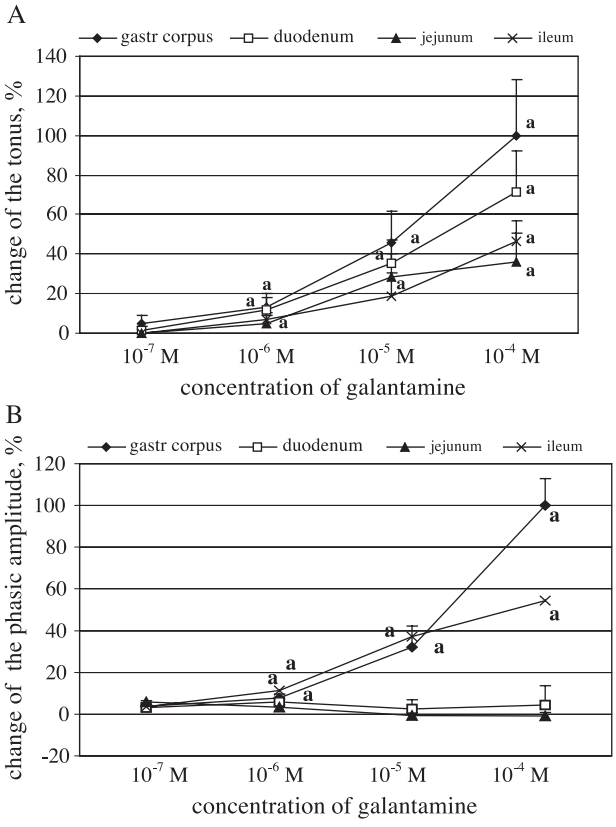

Fig. 3. Concentration-dependent curves of galantamine-induced effects on the smooth muscle preparations from rat gastrointestinal tract. (A) For the tonic contractions. The maximum contraction from 9.91 mN, caused by 10^{-4} mol/l galantamine on the gastric corpus smooth muscle strips was accepted as a 100% tonic contraction; (B) for the phasic reactions. The maximum amplitude alteration from 8.82 mN, caused by 10^{-4} mol/l galantamine on the gastric corpus smooth muscle strips was accepted as a 100% phasic reaction. The comparisons were made between reactions of different galantamine concentrations on every type smooth muscle preparation separately, [a]$P<0.05$.

3.2. Effect of galantamine on the spontaneous mechanical activity of isolated gastrointestinal smooth muscle samples

Galantamine influenced the spontaneous mechanical activity of isolated rat gastrointestinal smooth muscle preparations. The treatment of the samples with an increasing concentration of galantamine (1×10^{-7} to 1×10^{-4} mol/l) caused concentration-dependent tonic contractions. The maximal contractions were recorded 80 s–3 min after the treatment. The concentration-dependent curves of the galantamine-influence on the tonus of the smooth muscle preparations are illustrated in Fig. 3A. The anticholinesterase drug (at concentrations $\geq10^{-6}$ mol/l) significantly increased ($P<0.05$) the amplitude of spontaneous phasic activity of the smooth muscle preparations from gastric corpus and ileum. The jejunal smooth muscle samples demonstrated an opposite tendency. Galantamine has no effect on the amplitude of spontaneous phasic activity of duodenal strips. Those effects are shown in Fig. 3B. Galantamine-caused changes in the frequency of the phasic contractions were not significant on all kinds of smooth muscle preparations.

3.3. Influence of M-acetylcholine receptor antagonists on galantamine-induced smooth muscle reactions

In a concentration of 1×10^{-6} mol/l, the non-selective blocker of M-acetylcholine receptors atropine minimized significantly ($P<0.05$) galantamine (1×10^{-7}–1×10^{-4} mol/l)-provoked reactions in gastric corpus, duodenum, jejunum and ileum. It suppressed not only the tonic contractions, but also the amplitude of phasic contractions. The non-selective M-acetylcholine receptor antagonist ipratropium (1×10^{-6} mol/l) possessed similar, but weaker actions on all kinds of SM preparations. The influences described above are quantitatively presented in Table 2.

3.4. Influence of N-acetylcholine receptor antagonist hexametonium on galantamine-induced smooth muscle reactions

Hexametonium (1×10^{-6} mol/l) did not influence significantly the concentration-dependent augmentation of the tonus of all kinds of smooth muscle preparations, caused by galantamine (1×10^{-7} to 1×10^{-4} mol/l). The inhibitor of

Table 2
The galantamine-induced reactions in presence of 1×10^{-6} mol/l atropine and 1×10^{-6} mol/l ipratropium

Smooth muscle preparation	Antagonist	Concentrations of galantamine			
		10^{-7} mol/l	10^{-6} mol/l	10^{-5} mol/l	10^{-4} mol/l
Tonic contractions of smooth muscle preparations, caused by galantamine, %					
Gastric corpus	Control (9)	5.05±3.84	13.03±4.85	45.45±16.15	100.00±28.40
	Atropine (9)	0[a]	0[a]	7.10±4.04[a]	13.20±4.04[a]
	Ipratropium (6)	0[a]	5.17±2.03[a]	8.27±5.15[a]	19.18±3.03[a]
Duodenum	Control (9)	1.21±2.10	11.82±5.57	35.66±11.20	71.31±20.88
	Atropine (7)	0[a]	3.03±2.00[a]	7.06±3.03[a]	10.09±3.08[a]
	Ipratropium (6)	0[a]	5.55±3.12	8.27±3.32[a]	16.75±4.04[a]
Jejunum	Control (9)	0	4.94±2.34	28.66±11.50	35.82±14.53
	Atropine (9)	0	0[a]	4.03±3.03[a]	7.16±3.03[a]
	Ipratropium (6)	0	0[a]	7.45±4.14[a]	14.23±3.13[a]
Ileum	Control (9)	0	6.66±3.71	18.57±11.60	46.62±10.49
	Atropine (9)	0	0[a]	4.54±2.22[a]	7.06±3.43[a]
	Ipratropium (6)	0	2.05±1.28	6.16±3.04[a]	13.43±4.24[a]
Change of phasic amplitude of smooth muscle preparations, caused by galantamine, %					
Gastric corpus	Control (12)	3.74±0.90	7.82±1.81	32.27±9.98	100.00±12.77
	Atropine (9)	0[a]	3.06±1.25[a]	7.03±1.47[a]	13.85±2.35[a]
	Ipratropium (6)	0[a]	5.12±1.02	13.26±3.51[a]	22.47±2.27[a]
Duodenum	Control (9)	3.18±0.90	5.92±1.82	2.54±0.45	4.41±1.89
	Atropine (7)	0[a]	2.15±1.47	0.68±0.32[a]	0.35±0.32[a]
	Ipratropium (6)	0[a]	1.49±0.57[a]	1.81±1.02	2.33±1.04
Jejunum	Control (9)	5.78±1.70	3.41±1.36	−0.61±0.45	−0.93±0.48
	Atropine (6)	0[a]	0[a]	1.13±0.33[a]	1.82±0.45[a]
	Ipratropium (6)	0[a]	−1.14±0.55[a]	−2.27±0.91[a]	−2.84±1.76
Ileum	Control (9)	3.63±1.48	11.34±2.63	37.18±5.56	54.39±7.48
	Atropine (7)	0[a]	3.40±1.44[a]	5.49±1.70[a]	14.98±2.72[a]
	Ipratropium (6)	0[a]	7.16±2.04	9.65±2.27[a]	21.45±3.18[a]

Each value of the tonic effects was presented as % from the maximum tonic contraction 9.91 mN, caused by 10^{-4} mol/l galantamine on the gastric corpus smooth muscle preparations, accepted as a 100% contraction. The alteration of the amplitude of phasic contractions was presented as % from the maximum amplitude change from 8.82 mN, caused by 10^{-4} mol/l galantamine on the gastric corpus smooth muscle preparations, accepted as a 100% alteration. The sign "−" indicates a decrease of the amplitude. The numbers in brackets showed number of experiments. The galantamine reactions in presence of atropine or ipratropium were compared with control galantamine-induced reactions.

[a] $P<0.05$.

N-type acetylcholine receptors changed the amplitudes of galantamine-induced phasic reactions of the smooth muscle preparations from gastric corpus, jejunum and ileum: in gastric corpus and ileum, they were significantly reduced ($P<0.05$) and those of the jejunum were increased ($P<0.05$) as compared with relevant control reactions (Fig. 4).

3.5. Effects of methysergide on galantamine-induced mechanical activity

In the presence of 1×10^{-6} mol/l methysergide, there were differences in galantamine-induced reactions: a tendency for impairment of the tonic effect in comparison with control one in the all kinds of preparations was observed and a significant reduction in the amplitude of phasic contraction, caused by galantamine on ileal- and gastric corpus strips. The data are shown in Table 3.

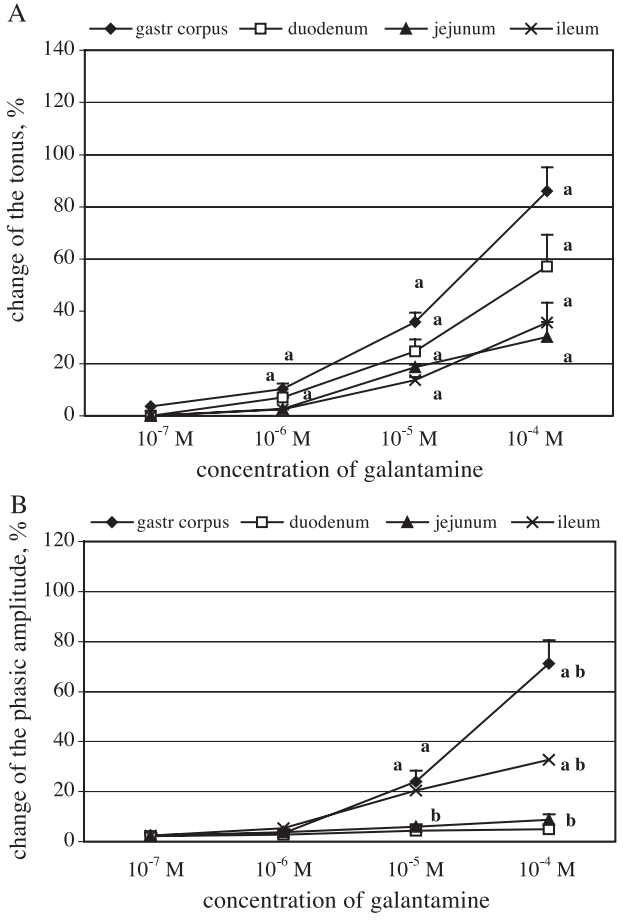

Fig. 4. Concentration-dependent curves of galantamine-induced effects on the smooth muscle preparations from rat gastrointestinal tract in presence of 1×10^{-6} mol/l hexametonium. (A) For the tonic contractions. Each value was presented as % from the maximum contraction from 9.91 mN, caused by 10^{-4} mol/l galantamine on the gastric corpus smooth muscle strips, accepted as a 100% tonic contraction; (B) for the phasic reactions. Each value was presented as % from the maximum amplitude change from 8.82 mN, caused by 10^{-4} mol/l galantamine on the gastric corpus smooth muscle strips, accepted as a 100% phasic reaction. The comparisons were made between reactions of different galantamine concentrations on every type smooth muscle preparation separately (A).The galantamine-induced effects in presence of hexametonium were comparison with the relevant control reactions also (B), [a], [b]$P<0.05$.

4. Discussion

The 20-day treatment with galantamine leads to functional disturbances in rat gastrointestinal tract. Galantamine-treated rats possess a gastrointestinal hyper tonus in comparison with control animals. It is well known that galantamine inhibits acetylcholinesterase and increases the level of endogenous acetylcholine. The latter interacts with M-acetylcholine receptors on smooth muscle cells. For that reason, the non-selective antagonist atropine and to a smaller degree ipratropium significantly block the galantamine effect on all kinds of smooth muscle preparations. The receptor activation is the cause for an increased intracellular Ca^{2+} level (Ogura, 2002) that leads to tonic contractions of smooth muscle preparations in concentration-dependent manner. These tonic reactions are not reduced significantly in the presence of hexametonium. This probably shows the unimportant participation of intramural neuronal N-acetylcholine receptors in their development.

Tonic effects relatively largest in size were observed in gastric and duodenal smooth muscle preparations. Most probably because of that, following galantamine application, the rats had well-visualized narrowed areas in their duodenum. These regions with lumen narrowing show locally increased tone and create resistance against moving of the enema into the lower levels of the tract. The increased resistance in the duodenum accounts for the lack of a contrast medium in the intestines of galantamine-treated rats immediately after probing. The increased resistance can also explain the delay of the contrast until the 3rd hour in the treated animals in comparison to the control rats. The increased tone in the stomach is relatively stronger than that in the duodenum and significantly exceeds the jejunal one. Therefore, the stomach contraction force permits the enema to overcome the narrow regions and with a some delay to reach the ileum.

A significant increase in the amplitude of phasic contractile activity is registered in ileal and stomach preparations only. In vitro indicated amplitude enhancement in the ileum is the cause for the intensification of peristaltic activity (Bollan, 1995), which we observed on that level of the gastrointestinal tract. It is the likely factor, which in a combination with the hyper tone of the gastrointestinal tract, ensures faster evacuation of the enema after the 3rd hour.

The variety of the present results suggests some differences in cholinergic control along rat gastrointestinal tract. It was established long ago that motility of the gastrointestinal tract is controlled by enteric inhibitory and excitatory motor neurons that innervate smooth muscle layers (Brookes, 1993). More than 30 functional types of neurons are present and about 25 different possible neurotransmitters have been

Table 3
The galantamine-induced reactions in the presence of 1×10^{-5} mol/l methysergide

Smooth muscle preparation	Antagonist	Concentrations of galantamine			
		10^{-7} mol/l	10^{-6} mol/l	10^{-5} mol/l	10^{-4} mol/l
Tonic contractions of smooth muscle preparations, caused by galantamine, %					
Gastric corpus	Control (9)	5.05±3.84	13.03±4.85	45.45±16.15	100.00±28.40
	Methysergide (8)	2.33±1.22	6.54±1.82	32.89±8.87	76.61±13.44
	Control (9)	1.21±2.10	11.82±8.57	35.66±11.20	71.31±20.88
Duodenum	Methysergide (7)	0.81±1.61	7.76±2.43	24.28±10.77	61.96±16.99
	Control (9)	0	4.94±4.34	28.66±11.50	35.82±14.53
Jejunum	Methysergide (7)	0	2.62±1.51	12.92±4.76	32.50±10.50
	Control (9)	0	6.66±3.71	18.57±11.60	46.62±10.49
Ileum	Methysergide (7)	0	3.32±0.81	14.73±3.83	34.41±7.61
Change of phasic amplitude of smooth muscle preparations, caused by galantamine, %					
Gastric corpus	Control (12)	3.74±0.90	7.82±1.81	32.27±9.98	100.00±12.77
	Methysergide (8)	1.70±0.56[a]	4.50±1.36[a]	24.15±7.03	61.38±11.10[a]
Duodenum	Control (9)	3.18±0.90	5.92±1.82	2.54±0.45	4.41±1.89
	Methysergide (7)	2.27±0.88	3.86±1.17	2.83±1.70	3.64±0.68
Jejunum	Control (9)	5.78±1.70	3.41±1.36	−0.61±0.45	−0.93±0.68
	Methysergide (7)	2.27±1.70	2.30±1.70	−1.36±0.68	−2.04±0.68
Ileum	Control (9)	3.63±1.48	11.34±2.63	37.18±5.56	54.39±7.48
	Methysergide (8)	2.83±1.81	3.64±2.04[a]	19.61±4.77[a]	28.19±6.24[a]

Each value of the tonic effects was presented as % from the maximum tonic contraction 9.91 mN, caused by 10^{-4} mol/l galantamine on the gastric corpus smooth muscle preparations, accepted as a 100% contraction. The alteration of the amplitude of phasic contractions was presented as % from the maximum amplitude change from 8.82 mN, caused by 10^{-4} mol/l galantamine on the gastric corpus smooth muscle preparations, accepted as a 100% alteration. The sign "−" indicates a decrease of the amplitude. The numbers in brackets after antagonists show number of experiments. The galantamine reactions in presence of methysergide were compare with the respective control galantamine-induced reactions.

[a] $P<0.05$.

identified in enteric neurons (McConalogue and Furness, 1994). In a significant part of them, the presence of different types M- and/or N-acetylcholine receptors were determined (Vizi et al., 1989). We consider that the enhanced level of endogenous acetylcholine, produced by acetylcholine inhibition, interacts with these receptors and stimulates the release of some neurotransmitter substances. The effect probably is additionally increased by the galantamine-provoked of N-acetylcholine receptors sensitization (Maelicke et al., 2001; Coyle and Kershaw, 2001). The participation of neuronal N-type acetylcholine receptors is proved (especially up to the phasic activity) in the presence of hexametonium by the decreased galantamine contractile efficiency.

Probable modulators of gastrointestinal motility following M- or N-neuronal acetylcholine receptor activation are 5-hydroxytriptamine (5-HT) (Ovsiannikov, 1999). Even 5-HT is accepted as a basic transmitter in neurons of the enteric ganglia (McConalogue and Furness, 1994). Immunohistochemical investigations in rat stomach, duodenum, jejunum, ileum, and colon demonstrate that 5-HT neurons are located in the myenteric- and submucous plexus (Fujimiya et al., 1997). There are 5-HT_3- and 5-HT_4-type receptors in the gastric and intestinal smooth muscle tissues. Some heterogeneity exists in their distribution along the gastrointestinal tract (Mine et al., 1997). There are no data for a possible direct effect of anticholinesterase drug on 5-HT-receptors. Significantly impairs in spite of all the blocking of 5-HT receptors with methysergide the galantamine-induced increase of the phasic amplitude of ileum and gastric corpus and did not influence significantly its tonic contractile effects. It means that the serotoninergic component significantly participates in the genesis of the galantamine-provoked phasic reaction of smooth muscle at these levels of the gastrointestinal tract. 5-HT that causes it is probably secreted from the intramural neuronal structures after the interaction of their M- and N-type acetylcholine receptors with endogenous acetylcholine. That conclusion is consistent with the fact that galantamine-induced augmentation of the spontaneous mechanical activity is tetrodotoxin-sensitive, whereas the contractile effect of the drug on the tone is not tetrodotoxin-sensitive (Mutafova-Yambolieva et al., 1993).

Galantamine induces a specific reaction in jejunal preparations: the increase of the drug concentration decreases the size of phasic amplitude and causes a significant tonic contraction. The tendency of the decrease of the phasic amplitude is kept in presence of ipratropium, it is not manifested on the background of atropine and is transformed into excitation process in the presence of hexametonium. The data indicate that the reaction is a result of N-acetylcholine receptor activation and occasionally of presynaptic M-acetylcholine receptors, which are weakly influenced by ipratropium and blocked by atropine. The stronger efficiency of atropine may be related to some specific influence on the presynaptic M-acetylcholine receptors. The reduction of the phasic amplitude is probably connected with the action of inhibitor substances, released

from the intramural neuronal structures at this level of the tract.

5. Conclusions

The application of galantamine (a single dose of 1 mg/kg per os) for 20 days increased rat gastrointestinal tonus, induced spastic reactions in the duodenum, increased peristaltic activity in the stomach and ileum and accelerated the evacuation kinetics.

Galantamine intensified in vitro the spontaneous mechanical activity of SM-tissues from rat gastrointestinal tract: cause concentration-dependent tonic contractions and augment the spontaneous contractile activity (without smooth muscle samples from jejunum and duodenum).

Galantamine-induced reactions had a complex genesis. They consist of two components: muscular, that is a result from the action of endogenous acetylcholine on M-type smooth muscle acetylcholine receptors and neuronal—from its influence on N- and M-type acetylcholine receptors within intramural neuronal structures. The muscular component caused tonic and phasic effects. The neuronal component induced significantly increase of amplitude of phasic contractions only. Whereas in smooth muscle tissues from gastric corpus and ileum, it was mainly brought about by 5-HT, probably secreted from intramural nerves. The influence on the amplitude of jejunal smooth muscle phasic contraction is affected by some inhibitory neurotransmitter substances.

In vivo and in vitro results show differences in the effects of galantamine on smooth muscle tissues at various levels of the gastrointestinal tract. They suggest certain qualitative differences in cholinergic control along rat gastrointestinal tract.

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References

Bollan, B, 1995. Calcium–contraction coupling in mouse urogenital smooth muscles. PhD thesis. Universite catolique de Louvain, pp. 14–15.

Brookes, S.J., 1993. Neuronal nitric oxide in the gut. J. Gastroenterol. Hepatol. 8 (6), 590–603.

Corey-Bloom, J., 2003. Galantamine: a review of its use in Alzheimer's disease and vascular dementia. Int. J. Clin. Pract. 57 (3), 219–223.

Coyle, J., Kershaw, P., 2001. Galantamine, a cholinesterase inhibitor that allosterically modulates nicotinic receptors: effects on the course of Alzheimer's disease. Biol. Psychiatry 49 (3), 289–299.

Cummings, J.L., 2003. Use of cholinesterase inhibitors in clinical practice: evidence-based recommendations. Am. J. Geriatr. Psychiatry 11 (2), 131–145.

Darvesh, S., Walsh, R., Kumar, R., Caines, A., Roberts, S., Magee, D., Rockwood, K., Martin, E., 2003. Inhibition of human cholinesterases by drugs used to treat Alzheimer disease. Alzheimer Dis. Assoc. Disord. 17 (2), 117–126.

Fujimiya, M., Okumiya, K., Yamane, T., Maeda, T., 1997. Distribution of serotonin-immunoreactive nerve cells and fibers in the rat gastrointestinal tract. Histochem. Cell Biol. 107 (2), 105–114.

Fulton, B., Benfield, P., 1996. Galanthamine. Drugs Aging 9 (1), 60–67.

Maelicke, A., 2000. Allosteric modulation of nicotinic receptors as a treatment strategy for Alzheimer's disease. Dement. Geriatr. Cogn. Disord. 11 (Suppl. 1), 11–18.

Maelicke, A., Samochocki, M., Jostock, R., Fehrenbacher, A., Ludwig, J., Albuquerque, E.X., Zerlin, M., 2001. Allosteric sensitization of nicotinic receptors by galantamine, a new treatment strategy for Alzheimer's disease. Biol. Psychiatry 49 (3), 279–288.

McConalogue, K., Furness, J.B., 1994. Gastrointestinal neurotransmitters. Bailliére's Clin. Endocrinol. Metab. 8 (1), 51–76.

Mine, Y., Yoshikawa, T., Oku, S., Nagai, R., Yoshida, N., Hosoki, K., 1997. Comparison of effect of mosapride citrate and existing 5-HT4 receptor agonists on gastrointestinal motility in vivo and in vitro. J. Pharmacol. Exp. Ther. 283 (3), 1000–1008.

Mutafova-Yambolieva, V.N., Yamboliev, I.A., Mihailova, D.N., 1993. Comparative effects of the anticholinesterase drug galanthamine on the mechanical activity of isolated rat jejunum and ileum. Gen. Pharmacol. 24 (5), 1253–1256.

Nordberg, A., Svensson, A.L., 1999. Cholinesterase inhibitors in the treatment of Alzheimer's disease: a comparison of tolerability and pharmacology. Drug Safety 20 (2), 146.

Ogura, T., 2002. Acetylcholine increases intracellular Ca^{2+} in taste cells via activation of muscarinic receptors. J. Neurophysiol. 87 (6), 2643–2649.

Ovsiannikov, V.I., 1999. Integrative mechanisms of the formation of the small intestine motor effects. Ross. Fiziol. Z. Im. I.M. Secenova 85 (9–10), 1278–1289.

Poirier, J., 2002. Evidence that the clinical effects of cholinesterase inhibitors are related to potency and targeting of action. Int. J. Clin. Pract., Suppl. 127, 6–19.

Scott, L.J., Goa, K.L., 2000. Galantamine: a review of its use in Alzheimer's disease. Drugs 60 (5), 1095–1122.

Sramek, J.J., Frackiewicz, E.J., Cutler, N.R., 2000. Review of acetylcholinesterase inhibitor galantamine. Expert Opin. Investig. Drugs 9 (10), 2393–2402.

Vizi, E.S., Kobayashi, O., Torocsik, A., Kinjo, M., Nagashima, H., Manabe, N., Goldiner, P.L., Potter, P.E., Foldes, F.F., 1989. Heterogeneity of presynaptic muscarinic receptors involved in modulation of transmitter release. Neuroscience 31 (1), 259–267.

Zarotsky, V., Sramek, J.J., Cutler, N.R., 2001. Galantamine hydrobromide: an agent for Alzheimer's disease. Am. J. Health-Syst. Pharm. 60 (5), 446–452.