Systemic oxytocin and vasopressin excite gastrointestinal motility through oxytocin receptor in rabbits

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Abstract The aim of the present study was to investigate the effect of systemic oxytocin (OT) and vasopressin (VP) on the motility of stomach and duodenum. Two plastic balloons made of condom were inserted into stomach and duodenum to monitor the change of mean pressure. Intravenous injection of OT (0.1–0.8 μg kg⁻¹) or VP (0.02–0.08 IU kg⁻¹) dose-dependently increased the stomach and duodenum pressure. Pretreatment of atosiban (1 μg kg⁻¹), the specific OT receptor (OTR) antagonist, attenuated the excitatory effect of OT or VP on the pressure of stomach and duodenum. Pretreatment of V1880 (1 μg kg⁻¹), the specific V1 receptor blocker, did not influence this effect. So we conclude that both of OT and VP injected systemically increased the gastric and duodenum motility via OTR.

Keywords gastrointestinal motility, oxytocin, receptor, vasopressin.

INTRODUCTION

Oxytocin (OT) is traditionally regarded as a hormone that involved in the parturition and milk ejection. In recent years, more and more evidence indicated that OT may play a role in the regulation of the function of gastrointestinal (GI) tract. Oxytocin and OT mRNA was found throughout the GI tract.² Although OT receptor (OTR) was not detected by indirect immunofluorescence,¹ there is no doubt that the OTR is present on the GI tract because the OTR mRNA was found on the human GI tissue.² A huge number of functional studies indicated that exogenous or endogenous OT regulated the GI motility,³⁻¹⁰ sensation¹¹,¹² and immune response to inflammation.¹³,¹⁴ Several studies reported that OT influence the gastric emptying and intestinal transit,³⁻⁹,¹⁰ but the results were controversial. The effect of OT on the motility of stomach and intestine has not been clearly illustrated. In this study, to study the effect of OT on the motility of GI tract, OT was systemically administrated in rabbits and change of mean pressure in stomach and duodenum was compared before and after OT administration.

Oxytocin and vasopressin (VP) are structurally-related cyclic nonapeptides that incorporate a disulfide bridge in their structures.¹⁵ Their sequences differ only in two of the nine amino acids.¹⁵ All of the currently known VP/OTR subtypes (V1a, V1b, V2, OT) have been cloned from rat and human tissues and characterized. They are members of the G protein-coupled superfamily of receptors and show significant structural homology to one another.¹⁵ The receptor selectivity of OT and VP for their own receptors is not absolute and significant cross-talk can occur with OT at VP receptors (and vice versa) at higher concentrations.¹⁵ Vasopressin may also participate in the regulation of GI motility.¹⁶ Although exogenous administration of VP influenced the GI motility, the receptor involved has not been clearly demonstrated. In this study, two specific OT/VP receptor antagonist, atosiban and V1880, was used to discriminate the subtype of receptors that mediate the effect of OT/VP on the motility of GI tract.

MATERIALS AND METHODS

Animal preparation

Male New Zealand white rabbits were provided by the animal centre of Shandong University. They were fasted overnight before the experiments. The treatment of animals was followed by the guidance of the Animal
Ethical Committee of the Medical School of Shandong University.

After anesthetized by urethane (1.0 g kg\(^{-1}\)), the trachea of the rabbit was cannulated to facilitate the ventilation. A plastic ballonet made of the condom (KL-1; Tianjin Human-Care Latex Cor., Tian Jin, China) was inserted into the stomach. Another end of the catheter was connected with a pressure transducer (VP-101; Xinhang Xingye Tech. Co., Beijing, China). A midline laparotomy was performed and another plastic ballonet was inserted into the duodenum through a hole made at the junction of the stomach and duodenum. Both of the ballonets were connected with the same transducer through a catheter. To monitor the blood pressure (BP), the right femoral artery was cannulated and the catheter was connected with the third transducer. The signals from the three transducers were separately amplified by three wave carrier amplifiers (BL-420; Taimeng Technical Cor., Chengdu, China) and recorded by a polygraph (BL-420; Taimeng Technical Cor.).

Three to 10 mL of normal saline was infused into the ballonet to maintain the pressure in the ballonet at about 10 mmHg. To stabilize the background motor after surgery, the gut was initially equilibrated for 30 min before any experiments were conducted.

Protocols

Experiment 1, dose-dependence of the effect of OT on BP, motility of stomach and duodenum.

Twenty-four rabbits were divided into four groups (group 1–4). Four doses of OT, 0.1, 0.2, 0.4, 0.8 g kg\(^{-1}\), were injected intravenously into these animals, with one dose in one group.

Experiment 2, pretreatment of atosiban or V1880 before the treatment of OT.

Two groups of rabbits with six in each was pretreated with atosiban [group 5] or V1880 [group 6] 5 min before the OT was administrated.

Experiment 3, similar with Experiment 1 except that OT is substituted by VP [group 7–10].

Experiment 4, similar with experiment 2 except that OT is substituted by VP [group 11–12].

Chemicals

Urethane and NaCl were bought from the Dongfeng Chemicals [Shanghai, China], OT [Cys-Tyr-Ile-Gln-Asn-Cys-Pro-Leu-Gly-NH\(_2\)], Arginine VP [Cys-Tyr-Phe-Gln-Asn-Cys-Pro-Arg-Gly-NH\(_2\)] and V1880 ([deamino-Pen1,O-Me-Tyr2,Arg8]-VR) are the products of Sigma [Saint Louis, MO, USA], atosiban [1-deamino-2-D-Tyr(OEt)-4-Thr-8-Orn-OT] was obtained from Ferring Company [Malmo, Sweden]. All chemicals were dissolved in normal saline frozen in aliquots that were thawed when required.

Statistical analysis

The mean value of the average (blood, stomach and duodenal) pressure for 60-s periods recorded over 0–3 min before treatment with OT or VP was taken as the baseline. In the groups pretreated with atosiban or V1880, the baseline was the average pressure over 2–3 min after the blockers were administrated. The average pressure for a 2- to 4-min period after each OT or VP treatment was normalized by subtracting the baseline, where the baseline for each experiment was equal to 0. This was taken as the change in pressure due to each treatment. The change of pressure was presented as mean ± SEM, with \(n\) indicating the number of rabbits. Data were analysed with SigmaStat 3.5 software (SPSS Inc., Chicago, IL, USA). One-way analysis of variance was used to analyse the effect of OT or VP on BP, mean pressure of stomach and duodenum. Mann–Whitney \(U\)-test was used to test the difference between the data of the group with the pretreatment of antagonist with that of the control. \(P < 0.05\) was considered to be significantly different.

RESULTS

Intravenous injection of OT and VP transiently increase the BP

Intravenous (i.v.) injection of OT (0.4 \(\mu g\) kg\(^{-1}\)) transiently increased BP [Fig. 1A]. One minute later after OT administration, the BP increased by 3.5 ± 1.0 mmHg (\(P < 0.05\), \(n = 6\)). Two minutes later, it returned to normal [Fig. 1]. Atosiban (1 \(\mu g\) kg\(^{-1}\), i.v.) did not influence the effect of OT on the BP, but V1880 (1 \(\mu g\) kg\(^{-1}\), i.v.), the specific antagonist of VP1 receptor, completely abolished the increase of BP followed by OT administration (\(P = 0.04\), \(n = 6\)).

Similar effect was observed after the administration of VP [Fig. 1B]. At 1 min, systemic VP [0.02 IU kg\(^{-1}\), i.v.] significantly increased BP by 9.4 ± 1.0 mmHg (\(P < 0.05\), \(n = 6\)). This effect was completely abolished by V1880 (1 \(\mu g\) kg\(^{-1}\), i.v.) and was not influenced by atosiban (1 \(\mu g\) kg\(^{-1}\), i.v.).

Effect of systemic OT on the GI motility

Intravenous injection of OT (0.2–0.8 \(\mu g\) kg\(^{-1}\)) dose-dependently increased the motility of stomach and
duodenum (Fig. 2A). This effect was observed 1 min after OT administration, reached the highest level at 3–5 min and return to normal 15 min later (Fig. 1). Three minutes after OT (0.6 µg kg$^{-1}$) administration, the mean pressure of stomach increased by

![Figure 1](image1)

**Figure 1** A. Represent recording of the effect of systemic oxytocin (OT) (0.4 µg kg$^{-1}$) on the blood pressure (upper trace), mean pressure stomach (middle trace) and duodenum (below) in rabbits. Arrow (↓), marker of OT administration. B. Represent recording of the effect of systemic vasopressin (VP) (0.04 IU kg$^{-1}$) on the blood pressure (upper trace), mean pressure of stomach (middle trace) and duodenum (below) in rabbits. Arrow (↓), marker of VP administration.

![Figure 2](image2)

**Figure 2** A. Dose-dependence of the systemic oxytocin (OT) on the motility of stomach and duodenum. The upper panel represents the change of mean intragastric pressure 3 min following different doses of OT administrated, lower panel is that of mean intraduodenal pressure. From 0.2 to 0.8 µg kg$^{-1}$, systemic OT significantly increases the mean pressure of stomach and duodenum. *P < 0.05 vs the data before OT administration. n = 6 in each group. B. Pharmacological manipulation of the effect of systemic OT (0.4 µg kg$^{-1}$) on the mean pressure of stomach and duodenum. The upper panel represents the change of mean gastric pressure and lower panel is that of the duodenum. In both of the two histograms, the first column is an increase of gastric cintestinal pressure 3 min following OT administration. The second and third column represents the data taken from animals with the pretreatment of atosiban (1 µg kg$^{-1}$) or V1880 (1 µg kg$^{-1}$). Note that pretreatment of atosiban blocks the excitatory effect of OT on the motility of stomach and duodenum, but V1880 does not influence it. n = 6 in each group.
11.0 ± 0.3 mmHg \( (P < 0.05, n = 6) \), that of duodenum increased by 8.6 ± 3.6 mmHg \( (P < 0.05, n = 6) \) \( \text{[Fig. 2A]} \).

Pretreatment of atosiban (1 µg kg\(^{-1}\)) completely abolished the excitatory effect of OT (0.4 µg kg\(^{-1}\), i.v.) on the motility of stomach and duodenum. With the treatment of atosiban, the change of mean pressure of stomach 3 min after OT administration is 2.9 ± 2.9 mmHg, that of duodenum is 0.8 ± 1.5 mmHg, significantly lower \( (P < 0.05, n = 6) \) than that of the group treated by the same dose of OT but without pretreatment of atosiban (10.8 ± 12 mmHg and 4.6 ± 1.8 respectively), which act as the control \( \text{[Fig. 2B]} \).

Pretreatment of V1880 (1 µg kg\(^{-1}\)) does not influence the excitatory effect of VP on the motility of stomach and duodenum \( \text{[Fig. 2B]} \).

**Effect of systemic VP on the GI motility**

Intravenous injection of VP \( (0.01–0.08 \text{ IU kg}^{-1}) \) dose-dependently increased the motility of stomach and duodenum \( \text{[Fig. 3A]} \). This excitatory effect appeared 1 min after VP administration, reached the highest level at 3 min and disappeared 10 min later \( \text{[Fig. 1B]} \). Three minutes after VP \( (0.04 \text{ IU kg}^{-1}, \text{i.v.) administration, the mean pressure of stomach increased by 1.1 ± 0.1 mmHg} \ (P < 0.05, n = 6), that of duodenum increased by 3.1 ± 0.7 mmHg \ (P < 0.05, n = 6) \ (\text{Fig. 3A)} \).

Atosiban completely abolish the excitatory effect of systemic VP \( (0.02 \text{ IU kg}^{-1}) \) on the motility of stomach and duodenum. In the group with the pretreatment of atosiban \( (1 \mu g \text{ kg}^{-1}, \text{i.v.)}, 3 \text{ min after VP administration, the change of mean stomach pressure is 0.3 ± 0.3 mmHg, that of duodenum is 0.5 ± 0.9 mmHg. Both of these were significantly lower than that of the control group} \ (1.1 ± 0.1 and 3.1 ± 0.7 mmHg \ (P < 0.05, n = 6) \ (\text{Fig. 3B)} \). Pretreatment of V1880 \ (1 \mu g \text{ kg}^{-1}, \text{i.v.)} \ did not influence the excitatory effect of VP on the motility of stomach and duodenum \ (P > 0.05, n = 6) \ (\text{Fig. 3B)} \).
Effect of atosiban and V1880 on BP, motility of stomach and duodenum

Intravenous injection of atosiban (1 \( \mu \text{g kg}^{-1} \), i.v.) and V1880 (1 \( \mu \text{g kg}^{-1} \), i.v.) did not influence the BP, motility of stomach and duodenum (data not shown).

DISCUSSION

The result of this study indicates that intravenous injection of OT or VP induced an increase of BP and elevation of gastric and duodenum pressure. The effect on cardiovascular system is abolished by V1880, the V1 receptor antagonist,\(^{17}\) and that on GI was attenuated by atosiban, the OTR antagonist.\(^{4}\)

Oxytocin has been shown to influence cardiovascular parameters in various ways. Acute administration increases the BP while chronic administration decreases it.\(^{18,19}\) The acute effect was mediated by OTR\(^{19}\) while the mechanism of the chronic one is unknown. Consistent with these studies, we also recorded a transient increase of BP after the acute systemic treatment of OT. But contrary to the result of Petersson et al.,\(^{19}\) we found that V1880 completely abolished the increase of BP caused by OT and VP while atosiban did not influence it. This result indicated that both of the two chemicals excited the cardiovascular system through V1 receptor, but not through OTR. This difference may be attributed to the different doses of the atosiban used by the two groups. With high concentration, atosiban could also bind the V1 receptor and inhibit the VP-induced second messenger, although this inhibition is much weaker than that induced by OT.\(^{20}\) The dose of atosiban used by Petersson was 1 mg kg\(^{-1}\), much higher than that used in this study (1 \( \mu \text{g kg}^{-1} \)). The OT-induced increase of BP was abolished by V1880 further supported our finding that systemic administration of OT transiently increased BP through binding on the V1 receptor.

Systemic administration of VP (0.02 IU kg\(^{-1}\), i.v.) increased BP. Pretreatment of V1880 (1 \( \mu \text{g kg}^{-1} \)) completely abolished this effect, but the same dose of atosiban does not influence it. This is consistent with the commonly accepted theory that VP constricts blood vessels through V1a receptors.\(^{21,22}\) This result also indicate that the dose of V1880 (1 \( \mu \text{g kg}^{-1} \)) used in this study is sufficient to antagonize the effect of exogenous VP (0.02 IU kg\(^{-1}\)) on peripheral organs through V1 receptor.

Systemic OT increased the gastric and duodenum pressure. The increase of mean gastric pressure was mainly attributed to the increase of the phasic contraction of the stomach, both the strength and the frequency. So we believe that the motility of gastric body and antrum, mainly the circular muscle, was increased following systemic administration of OT. This result seems inconsistent with our previous studies which indicated that OT decreased the gastric emptying and intestinal transit in rats.\(^{3,9,10}\) One of the possibilities that could be used to explain this inconsistence is that there may be species difference concerning the mechanism of OT effect on GI tract. In rats, systemic administration of OT induced release of CCK, and the inhibitory effect of OT on gastric emptying and intestinal transit is mediated by CCK receptors.\(^{3,10}\) In this study, it seems that the excitatory effect of OT on the stomach and duodenum is mediated by OTR.

Endogenous OT may exert some physiological effects on the GI motility.\(^{4,8}\) In this study, we failed to record the change of GI motility following the treatment of atosiban. This data seems inconsistency with that of Ohlsson et al.,\(^{4}\) which indicate that atosiban delayed gastric emptying of a semisolid meal compared to saline in human. One of the possibilities that explain this inconsistence is that OT may have different effect at different period of the stomach. Oxytocin is released after a meal,\(^{23}\) so it seems that endogenous OT facilitate the mechanical rubbing of the food in stomach and increase gastric emptying after the ingestion of the food. Oxytocin may not exert influence on the GI motility during the fasting state.

Vasopressin synthesis in GI tract was also reported,\(^{24}\) but the effect on GI motility has not been illustrated. In this study, systemic administration of VP significantly increased the motility of stomach and duodenum. This effect was not influenced by pretreatment of V1880, which completely abolished the effect on cardiovascular system by the same agent. The excitatory effect of VP was abolished by atosiban, so it is possible that exogenous VP excite the GI motility mainly through the OTR.

In conclusion, the present study indicated that systemic administration of OT or VP excites cardiovascular system via V1 receptors and increase the GI motility through OTR.

ACKNOWLEDGMENTS

This study is supported by the research project of the Education Institute of Shandong Province [J05L17], the Natural Scientific Foundation of China (NSFC, No 30570832), the Natural Scientific Foundation of Shandong Province [Q05C01], and the ‘1020’ project of the Health Institute of Shandong Province.
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