EFFECT OF A NEW ANTICHOLINERGIC AGENT (SA-504) ON MOTILITY OF THE GASTROINTESTINAL TRACT AND THE URINARY BLADDER IN CATS

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Abstract—Effects of 1, 1-dimethyl-5-methoxy-3-(dithien-2-ylmethylene) piperidinium bromide (SA-504) on the motility of gastrointestinal tract and of the urinary bladder were examined in cats. The inhibitory activity of SA-504 on the spontaneous motility of the stomach brought about by i.v. injection was nearly equal to that of hyoscine-N-butylbromide (HB), but approx. one-fourth of atropine sulfate (Atr). Intragastric administration of either SA-504 or HB produced obvious inhibition on the spontaneous motility of the stomach, while their potencies were less than one-tenth of Atr. The spasmolytic activity of SA-504 on the stomach contraction induced by vagal stimulation was almost equal to that of HB and approx. one-fourth that of Atr. In contrast to the effect on spontaneous motility of the stomach, all these compounds exhibited a somewhat weaker inhibitory action on the contraction of the stomach induced by stimulation. Both SA-504 and HB exhibited more potent inhibition on spontaneous movements of the jejunum and the sphincter of Oddi than on the stomach. On the contrary, the inhibitory potencies of Atr on the jejunum and the sphincter of Oddi did not differ much from those observed in the stomach. The inhibitory effects of these compounds on the contraction of the colon induced by pelvic nerve stimulation was similar to that in the case of stomach contraction induced by vagal stimulation. Inhibition of SA-504 on the spontaneous motility of the urinary bladder was weak and hardly any effect was observed even at a dose inhibiting the gastric motility. HB inhibited the motility of the urinary bladder under the influence of the same dose as that reduced the gastric motility. On the other hand, a significant effect was observed with Atr, the activity of which was as strong as on the motility of the stomach. Blockade by these compounds of the contraction of the urinary bladder induced by pelvic nerve stimulation required much higher doses than those required for reduction of spontaneous movements of the bladder. Among the compounds tested, the activity of Atr on the contraction of the urinary bladder was the weakest.

1, 1-Dimethyl-5-methoxy-3-(dithien-2-ylmethylene) piperidinium bromide (SA-504) is a new compound having strong anticholinergic activity (1). In a previous report (2), it was demonstrated that the in vitro anticholinergic activity of SA-504 was approx. the same as that of atropine sulfate (Atr). In vivo experiments with rats revealed that inhibition by SA-504 of the stomach contraction induced by vagal stimulation was more marked than Atr. On the other hand, the antisialagogue and mydriatic activities as well as the inhibitory effect on gastric secretion of SA-504 were found to be weaker than that of Atr in rats or mice.
The present paper describes the effect of SA-504 on the motility of various regions of gastrointestinal tract, and the urinary bladder in anesthetized cats. The results have been compared to those of other known anticholinergic agents.

MATERIALS AND METHODS

Adult cats of both sexes weighing 2.2 to 3.5 kg, were used. Animals, fasted for 24 hr before the experiments, were anesthetized with ether and then with a chloralose-urethane mixture (weight ratio 1:5, 200-400 mg/kg i.v.). Drugs tested were SA-504, hyoscine-N-butylbromide (HB) and Atr. All drugs were dissolved in saline. In most experiments, the drug solution was injected into the femoral vein through the polyethylene cannula. The chemical structure of SA-504 is shown in Fig. 1.

Motility of the stomach

Spontaneous motility of the stomach was examined by recording the pressure change according to the balloon method of Perret et al. (3). A balloon attached to the polyethylene tube connected to a pressure transducer was introduced into the stomach through the oral cavity, and, thereafter, air was admitted into the balloon. In experiments of vagal stimulation, the balloon was fixed to the pyloric region through a midline abdominal incision. A uniform brief contraction of the stomach was induced by electrical stimulation (1-3 V, 2 msec duration, 10/sec for 5 sec) of the cervical vagus at 2 min intervals. In some experiments, drugs were administered directly into the stomach through the polyethylene cannula attached to the tip of the balloon, as a model for oral administration.

Motility of the jejunum and the sphincter of Oddi

Simultaneous measurements of spontaneous motility of the jejunum and the sphincter of Oddi were carried out in the same animal. According to the method of Liedberg et al. (4), the motility of the sphincter of Oddi was observed by recording the pressure change. A cannula connected to a pressure transducer was inserted into the common bile duct towards the duodenum. Saline was perfused through the cannula at a constant rate of 4.2 ml/min and motility of the sphincter was estimated by measuring the change in perfusion pressure. Motility of the jejunum was measured by the balloon method described above.
Motility of the colon

A water-filled balloon attached to a polyethylene tube was introduced into the descending colon in the direction of the anus. A brief contraction of the colon was evoked at 5 min intervals by electrical stimulation (2–5 V, 1 msec duration, 20/sec for 10 sec) of the pelvic nerve supplying the wall of the colon and the bladder. Changes in intraluminal pressure of the colon were examined by means of a pressure transducer and were recorded simultaneously with the gastric motility.

Motility of the urinary bladder

The urinary bladder was cannulated with a polyethylene tube through the urethra and distended to some degree by infusion of warm saline. Spontaneous motility of the urinary bladder was recorded by means of a pressure transducer. A uniform brief contraction was induced at 5 min intervals by electrical stimulation (1–3 V, 1 msec duration, 10/sec for 10 sec) of the branch of pelvic nerve supplying the wall of bladder.

RESULTS

Motility of the stomach

Regarding spontaneous motility of the stomach, SA-504, HB and Atr produced inhibitory responses characterized by a fall of the tone and reduction in the amplitude of peristaltic movements. Table 1 summarizes effects of drugs on spontaneous motility in cats. As illustrated in Table 1, inhibition of the amplitude of peristaltic movements and the duration of drug action were shown to be dose-dependent. The activity of SA-504, which inhibits movements by approx. 50% at a dose of 20 μg/kg i.v., was nearly equal to that of HB, but approx. one-fourth that of Atr.

When administered into the stomach, SA-504 effectively blocked the peristaltic movements at a dose of 2 mg/kg, as shown in Fig. 2. The potency of HB was approx. the same as that of SA-504, whereas Atr was more potent than SA-504. Regarding the size of pupil

| Drugs | Doses (μg/kg i.v.) | No. of expts. | Per cent inhibition* | Duration (min) |
|-------|-------------------|---------------|----------------------|----------------|
| SA-504| 20                | 10            | 47±3.5               | 11±1.1         |
|       | 40                | 9             | 69±6.9               | 17±2.5         |
|       | 80                | 6             | 83±9.8               | 23±1.9         |
| HB    | 20                | 8             | 39±6.5               | 8±1.3          |
|       | 40                | 11            | 68±1.9               | 13±1.7         |
|       | 80                | 4             | 74±7.9               | 20±1.0         |
| Atr   | 5                 | 10            | 44±5.6               | 15±2.0         |
|       | 10                | 9             | 73±5.6               | 30±3.0         |
|       | 20                | 3             | 92±3.3               | 53±4.4         |

Values are the means±standard error.

* amplitude of peristalsis
measured simultaneously with the gastric movements, 2 mg/kg of SA-504 and HB exerted no influence, but 0.2 mg/kg of Atr produced mydriasis.

Fig. 3 shows the inhibitory effects of drugs on the contraction induced by vagal stimulation. Result of 4 experiments.

Fig. 2. Inhibitory effects of SA-504 and other anticholinergics on spontaneous motility of the stomach. Drugs were administered orally. The values are the mean of 3 cats. —○— SA-504 2 mg/kg, —△— HB 2 mg/kg, —■— Atr 0.2 mg/kg.

Fig. 3. Effects of SA-504 and other anticholinergics on contraction of the stomach induced by vagal stimulation. I.V. injection of 40 μg/kg of SA-504 resulted in approx. 50% inhibition on the
TABLE 2. Inhibitory effects of SA-504 and other anticholinergics on spontaneous motility of the jejunum and the sphincter of Oddi.

| Drugs | Doses (μg/kg i.v.) | No. of expts. | Jejunum | Sphincter of Oddi |
|-------|--------------------|---------------|---------|-------------------|
|       |                    |               | Per cent inhibition* | Duration (min) | Per cent inhibition** | Duration (min) |
| SA-504 | 5                  | 6             | 87±4.3  | 11±1.9           | 17±7.5       | 7±1.0          |
|        | 20                 | 7             | 98±1.5  | 13±2.4           | 30±3.2       | 9±1.1          |
| HB     | 5                  | 7             | 91±3.6  | 8±0.7            | 18±3.5       | 6±0.8          |
|        | 20                 | 6             | 99±0.5  | 10±1.5           | 26±4.1       | 8±0.7          |
| Atr    | 5                  | 7             | 88±4.1  | 17±2.6           | 13±2.5       | 9±1.6          |
|        | 20                 | 4             | 97±2.1  | 28±3.4           | 20±5.6       | 18±2.7         |

Values are the means±standard error.

* amplitude of peristalsis

** perfusion pressure measured at the choledochoduodenal junction
contraction. The activity of HB was almost equal to that of SA-504, while Atr was approx. four times as potent as SA-504. At equipotent doses, Atr exhibited a much longer duration of action than SA-504 and HB.

Motility of the jejunum and the sphincter of Oddi

As illustrated in Table 2, a small amount of the test compounds (5 \( \mu \)g/kg i.v.) produced strong inhibition on the spontaneous movements of the jejunum. As in the case of the stomach, inhibitory effects were characterized by decrease in the amplitude of peristaltic movements and by inhibition of the tonicity in the smooth muscle of the jejunum. Inhibitory activity to the jejunum, however, was greater than that to the stomach in the case of each drug.

All three drugs also exhibited much the same inhibitory action on the spontaneous motility of the sphincter of Oddi as on the movements of the jejunum, though the action of Atr was longer lasting than that of SA-504 or HB (Table 2 and Fig. 4). It was also observed that these compounds had no influence on the arterial blood pressure during the course of the experiments.

![Graph showing effects of SA-504 and other anticholinergics on spontaneous motility of the stomach and on contraction of the colon induced by pelvic nerve stimulation.](image)

Fig. 5. Effects of SA-504 and other anticholinergics on spontaneous motility of the stomach and on contraction of the colon induced by pelvic nerve stimulation. Result of 4 experiments. Top: spontaneous motility of the stomach. Bottom: contraction of the colon induced by stimulation.
Motility of the colon

Fig. 5 represents the effects of drugs on the contraction of the colon elicited by electrical stimulation of the pelvic nerve. Forty μg/kg i.v. of SA-504 and HB inhibited the contraction by approx. 30% that of the control, while the inhibition caused by Atr was approx. 50% at a dose of 10 μg/kg i.v. It was further observed that the duration of action of Atr was obviously longer than that of SA-504 or HB. When the gastric motility was measured at the same time as the contraction of the colon, the maximum inhibitory effects of these drugs on the colon always lagged behind those on the stomach by a few minutes.

Motility of the urinary bladder

As shown in Fig. 6, SA-504 did not show any effect on the spontaneous motility of the urinary bladder at a dose of 20 μg/kg i.v., which produced a 50% inhibition on the peristaltic movements of the stomach. Even at 40 μg/kg i.v., SA-504 was less active as compared with other drugs. At a dose of 40 μg/kg i.v., HB clearly reduced the movements,
while a significant inhibitory effect was observed with 10 μg/kg i.v. of Atr on the motility of the urinary bladder as well as on the spontaneous gastric motility.

In contrast to the effect on the spontaneous motility, much higher doses of each drug were required to cause inhibition on the contraction induced by electrical stimulation of the pelvic nerve. As shown in Fig. 7, 1 mg/kg i.v. of SA-504 produced approx. 40% inhibition on the contraction of the urinary bladder. HB was almost equipotent to SA-504. On the contrary, Atr showed no significant influence at a dose of 1 mg/kg i.v. Even at a dose as high as 3 mg/kg i.v., at which dose the contraction was markedly inhibited by SA-504 and HB, Atr produced only a slight reduction in the contraction of the urinary bladder.

DISCUSSION

SA-504, a quaternary ammonium compound of piperidine derivatives, possesses strong anticholinergic activity (1, 2). It also exhibits potent spasmylytic action, which is mainly ascribable to the antimuscarinic activity (2), on the gastrointestinal tract after i.v. administration in rats. In general, the synthetic antispasmodic with quaternary ammonium nitrogen is known to be poorly absorbed through the digestive tract as compared with the tertiary amine compound (5). In the present experiments, however, it was demonstrated that SA-504 produced clear inhibition on the gastric motility when administered into the stomach, although the inhibitory action was obviously weaker than the effect of
an i.v. injection. Thirty to 60 min after intragastric administration of 2 mg/kg of SA-504, approx. a 60% inhibition was observed which lasted for more than 2 hr. This result indicates that a sufficient amount of the compound for reducing the gastric motility is well absorbed through the gastrointestinal tract. The fact that 14C-labelled SA-504 is absorbed through the digestive tract after oral administration to rats supports the above findings (6).

Regarding spontaneous motility of various areas of the gastrointestinal tract, SA-504 administered i.v. exhibited a more potent blockade of the jejunum and the sphincter of Oddi than the stomach. However, the compound produced no influence on the spontaneous motility of the urinary bladder at a dose which inhibits gastric movements. The effect of HB on the gastrointestinal motility was comparable to that of SA-504, but, in contrast to SA-504, HB inhibited motility of the urinary bladder under the exact dosage as that which had reduced the gastric motility. On the contrary, inhibitory potencies of Atr on the spontaneous motility of the jejunum, sphincter of Oddi and the urinary bladder did not differ much from the effect on the stomach as compared with SA-504 and HB.

As to the gastrointestinal contraction induced by parasympathetic nerve stimulation, it was found that the inhibitory effects of these compounds on the contraction of the colon were similar to those in the case of the stomach contraction. In addition, inhibition by these compounds was shown to be more potent in the spontaneous motility than in the contractile movements evoked by stimulation.

It was further observed that the blockade by each drug of the contraction of the urinary bladder evoked by pelvic nerve stimulation was much weaker than the inhibition on the gastrointestinal contraction induced by stimulation or on the spontaneous motion of the urinary bladder. Thus, even at a dose as high as 1 mg/kg i.v., SA-504 and HB inhibited the contraction of the urinary bladder only by approx. 40% that of the control. Atr, which strongly inhibited spontaneous motility of the urinary bladder, caused a very slight reduction of the contraction after an i.v. administration of 3 mg/kg.

In addition to these effects, Atr produced mydriasis but SA-504 and HB showed no mydriatic activity, at a dose which inhibits the spontaneous motility of the stomach after intragastric administration.

These findings indicate that the effect of each compound varies depending on the organ and/or the type of movements (spontaneous or evoked).

As reported in the previous paper (2), inhibition by SA-504 of the stomach contraction induced by vagal stimulation in the rat was 3 times as potent as that of Atr, whereas the activity of SA-504 in the cat was approx. one-fourth that of Atr, as examined in the present experiments. Therefore, spasmolytic action of the drug appears to be species-dependent.

It has been reported that response to parasympathetic nerve stimulation of various smooth muscle organs was not completely inhibited by Atr (7). Goldenberg et al. suggested that a non-cholinergic transmitter may be responsible for this partial atropine-resistant effect in the gastrointestinal tract (8). On the other hand, it is assumed that the
pelvic nerve terminal in the urinary bladder has a property like ganglion, since the dose required for reduction of the contraction of the urinary bladder induced by parasympathetic nerve stimulation is almost equal to the dose which inhibits the nictitating membrane contraction induced by electrical stimulation of the preganglionic fiber (9). In addition, it is suggested that a substance other than acetylcholine may be the transmitter substance released by the excitatory innervation of the urinary bladder (10-13). These possibilities may partly be responsible for the organ specificity of drugs or the difference in activity between the spontaneous and evoked movements observed in the present experiments.

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