Centrally Applied Bombesin Increases Nerve Activity of Both Sympathetic and Adrenal Branch of the Splanchnic Nerves

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ABSTRACT—We reported that centrally applied bombesin probably excites both the gastric sympathetic and adrenomedullary systems and thus induces inhibition of gastric acid secretion. In the present study, therefore, we examined whether or not centrally applied bombesin directly affects sympathetic nerve activities in rats anesthetized with urethane. Intracerebroventricular administration of bombesin (0.3 and 3.0 nmol) increased discharge rates of the sympathetic branch as well as those of the adrenal branch of preganglionic greater splanchnic nerves. These effects were not secondary to changes in arterial blood pressure by bombesin. In conclusion, centrally applied bombesin directly activates both the sympathetic and adrenomedullary systems.

Keywords: Bombesin, Sympathetic nerve activity, Adrenal nerve activity

Bombesin given intracerebroventricularly (i.c.v.) produces a marked increase in the plasma level of adrenaline, while that of noradrenaline is slightly increased only at relatively large doses of the drug (1–3). As related to possible roles of bombesin in central regulation of gastric functions, bombesin is known to inhibit gastric acid secretion (4). In our previous study, we suggested that centrally applied bombesin probably excites both the gastric sympathetic nerves and adrenomedullary system and thus induces inhibition of gastric acid secretion, on the basis of following observations (5): The inhibition of gastric acid secretion induced by i.c.v. applied bombesin was not modified either by chemical sympathectomy with 6-hydroxydopamine or by bilateral adrenalectomy alone. However, this inhibition by bombesin was completely abolished by bilateral adrenalectomy plus chemical sympathectomy with 6-hydroxydopamine as well as by bilateral cutting of the greater splanchnic nerves.

There is, however, no direct evidence of the effects of bombesin (i.c.v.) on the splanchnic sympathetic nerves, except for a report by Somiya and Tonoue (6). Furthermore, the results reported by them were not conclusive; the adrenal sympathetic nerve was either stimulated or attenuated by bombesin.

Preganglionic greater splanchnic nerves ramify into two major branches: the adrenal branch and the sympathetic branch terminating in the coeliac ganglion. Therefore, in the present study, we examined whether or not centrally applied bombesin directly affects the nerve activity of the sympathetic nerves and the adrenal branches of the preganglionic greater splanchnic nerves in rats anesthetized with urethane.

Male Wistar rats weighing 400–450 g were maintained in a room at 22–24°C under a constant day-night rhythm for 7–10 days and given food (laboratory chow, CE-2; Clea Co., Tokyo) and tap water ad libitum. Experiments were performed under urethane anesthesia (1.1 g/kg, i.p.). The femoral vein, the femoral artery and the trachea were cannulated for administration of drugs, arterial pressure recording and artificial respiration, respectively. The animal was then paralyzed with d-tubocurarine (1.5 mg/kg, i.v.) and artificially ventilated. The body temperature was monitored with a rectal thermometer and maintained at 36–37°C by an automatically regulated heating pad. Animals were placed in a stereotaxic apparatus. Bombesin dissolved in saline was given into the lateral cerebral ventricle (AP: 7.5, L: 1.1, H: 3.5 below the surface of the brain) in a volume of 10 µl through a stainless steel micropipette (0.35-mm outer diameter).

One of the right adrenal branch or the sympathetic branch of the preganglionic greater splanchnic nerve was exposed through a right retroperitoneal flank incision. The central cut end of the nerve was placed on a pair of stainless steel wire electrodes. The entire preparation was immersed in a mixture of liquid paraffin and vaseline in order to provide electrical insulation and secure a good
recording of efferent nerve activity over several hours.

The efferent nerve activity was amplified by a bio-
electric differential amplifier (model AB600G; Nihon
Kohden, Tokyo) (band path 100–1000 Hz, –3 dB) and
displayed on an oscilloscope (model 2G51; San-Ei Instru-
ment Co., Ltd., Tokyo) and fed into the real-time UNIX
signal processing computer (model 6450; Concurrent,
Tinton Falls, NJ, USA). Nerve activity was analyzed and
converted to the discharge rate by means of a software
package “Laboratory Workbench” (Concurrent). The
threshold level of the integration was set above the back-
ground noise level observed at postmortem.

Drugs used were: bombesin (Peptide Institute, Inc.,
Osaka); d-tubocurarine chloride, phenylephrine hydro-
chloride (Sigma Chemical Co., St. Louis, MO, USA);
sodium nitroprusside (Nacalai Tesque, Inc., Kyoto).
Results are expressed as means±S.E. Dunnett’s test
was used for multiple comparison after one-way analysis
of variance (ANOVA). Significant values are those with
P < 0.05.

As characteristically shown in Fig. 1, intravenous
administration of sodium nitroprusside (100 μg/kg), a
vasodilator, reduced arterial blood pressure and markedly
increased discharge rates of the adrenal branch of the
greater splanchnic nerve. On the other hand, intravenous
administration of phenylephrine (80 μg/kg), an adrener-
gic α-adrenoceptor agonist, elevated arterial blood pres-
sure and markedly suppressed discharge rates. Similar
responses by administration of nitroprusside and phenyl-
ephrine were also observed in the nerve activities of the
sympathetic branch. These results indicate that the
splanchnic nerve activities were negatively affected by
changes in arterial blood pressure.

Bombesin (0.3 and 3.0 nmol) applied i.c.v. induced a
marked and long lasting (at least 2 hr) increase in arterial
blood pressure. The maximal response was observed
between 20 and 80 min after the administration of this
peptide (3 nmol), and the mean maximal increment was
35.4±4.8 mmHg (n=7). As shown in Fig. 2, adminis-
tration of bombesin, i.c.v. in a dose of 3 nmol caused
marked and long lasting increases in discharge rates of the
sympathetic branch as well as that of the adrenal branch
of the greater splanchnic nerves. These long lasting in-
creases in discharge rates were correlated with those in
blood pressure. On the other hand, intravenous applica-
tion of bombesin at 3 nmol did not affect the discharge
rates of the sympathetic and the adrenal branches and
mean arterial blood pressure (data not shown). The
increment of discharge rates in both branches reached
a plateau about 90–150 min after the administration of
bombesin, and this was followed by a gradual recovery.
Discharge rates of the adrenal branch at 5, 30 and 90 min
after the administration were 214±44, 320±23 and
546±96 percent of the preadministration values, respec-
tively, while the values in the sympathetic branch were
184±20, 239±22 and 458±29, respectively. These effects
of bombesin in inducing increased nerve activities of both
branches were dose-dependent as shown in Fig. 3.

Circulating plasma catecholamines are thought to
reflect sympathoadrenal activity (7, 8). In contrast to
adrenaline, circulating plasma noradrenaline concentra-
tion can be elevated not only by an increased sympathetic
erve activity but also by an increased output of
noradrenaline from the adrenal glands (9, 10). It is there-
fore likely that an increase in plasma level of noradrena-
line is not always an index of activation of the sympa-
thetic nerves.

As shown in Fig. 1, perception of a decrease in arterial
blood pressure increased discharge rates of the adrenal
branch, as a homeostatic manifestation. However, cen-
trally applied bombesin increased the arterial blood pres-
sure accompanied by parallel increases in discharge rates
of the sympathetic branch as well as that of the adrenal
branch of preganglionic greater splanchnic nerves. It is
therefore evident that centrally applied bombesin direct-
ly, but not secondarily, affects discharge rates of both
sympathetic and adrenal branches of splanchnic nerves.
The present results support our previous indirect evidence
that centrally applied bombesin probably excites both the
gastric sympathetic nerves and adrenomedullary system
and thus induces inhibition of gastric acid secretion (5).

Accumulating evidence suggests that there exist
regional differences in the activities of sympathetic nerves
in response to bombesin and some other neuroactive sub-
stances: decreased sympathetic outflow in the interscapu-
lar brown fat by bombesin, i.c.v. (11, 12), and reduced
Fig. 2. The effect of bombesin (3.0 nmol, i.c.v.) applied on the discharge rates and action potentials of the adrenal (left panel) and the sympathetic branches (right panel) of the preganglionic greater splanchnic nerve. The original action potentials were drawn with a high speed thermal recorder.

Fig. 3. Effects of bombesin i.c.v. applied on the discharge rates of the adrenal and the sympathetic branches of the preganglionic greater splanchnic nerve. Values are expressed as a percentage of preadministration values. ○: Vehicle (Saline); ▲: Bombesin, 0.3 nmol; ■: Bombesin, 3.0 nmol. Numbers in parentheses represent the number of experiments. *P < 0.05, statistically significant difference from the respective control value with vehicle.
renal sympathetic nerve activity and increased cutaneous sympathetic nerve activity by prostaglandin E2, i.c.v. (13). Furthermore, i.v. administration of interleukin-1β increased the activity of the adrenal and the splenic nerves, and it induced a transient increase followed by a long-lasting suppression in the renal nerve activity (14). Effects of central bombesin on the sympathetic neuron system in the other regions are therefore the subject of ongoing investigations.

In the present study, we demonstrate a direct evidence that bombesin centrally activates sympathetic nerves within the splanchnic regions as well as adrenal medullas.

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