25 °C, and minimal or absent at 35 °C. The off-CO_2 transient hyperpnea is preceeded during CO_2 inhalation by inhibition of breathing f. At 15 °C, the lower metabolic rate and lower O_2 requirements of *Coluber* may faciliate this inhibition. At the higher Tbs, a higher metabolic rate and O_2-related ventilatory drive may prevent IPC-mediated inhibition from dominating ventilation during CO_2 breathing, thus minimizing the off-CO_2 transient increase in ventilation over the steady-state response level.

P. scripta showed no sign of ventilatory inhibition during inhalation of CO_2 at concentrations up to 6% and exhibited no off-CO_2 transient hyperpnea at any Tb[3]. The difference in the Tb dependence of the steady-state CO_2 sensitivities between *Coluber* and *Pseudemys* may reside in the presence in *Coluber* but not in *Pseudemys* of IPC-mediated ventilatory inhibition which is prominent at low but not high body temperatures.

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Parasympathetically evoked parotid salivary secretion of chronically amitriptyline-treated rats

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Summary. Treatment of rats with amitriptyline for 4 weeks significantly decreased flow rate of saliva elicited from parotid glands in response to electrical stimulation of the parasympathetic innervation but did not alter calcium concentration of such saliva. The mechanism of the dissociation between flow rate and calcium concentration of parasympathetically evoked saliva induced by amitriptyline treatment remains to be explored, and may not involve an amitriptyline induced reduction in acetylcholine release.

Tricyclic antidepressant drugs such as amitriptyline have been widely used in treatment of depression. However, one of the side effects following longterm treatment with antidepressants is hyposecretion or no secretion at all from the salivary glands[2-4]. It has been suggested but not established that the reduced salivary secretion is the result of anticholinergic effects of the drug[5].

The present study was undertaken therefore to determine if salivary secretion evoked by stimulation of the parasympathetic innervation to parotid is reduced in rats chronically treated with amitriptyline. Furthermore, since calcium concentration of parasympathetically evoked saliva is related to salivary flow rate[6], the effects of amitriptyline on calcium concentration were also examined; the main objective in this case was to see if a drug-induced separation of calcium concentration from flow rate could be effected.

Materials and methods. Female Long-Evans rats, 4-6 months old, and weighing between 200 and 250 g were used in these experiments. Rats were administered daily 10 mg/kg of amitriptyline i.p. for 2 or 4 weeks. The experimental and control rats were maintained on rat lab chow and water ad libitum until 18 h before the experiments, when food but not water was removed. The rats were anesthetized by i.p. administration of sodium pentobarbital (50 mg/kg b.wt). The parasympathetic innervation to the parotid gland was stimulated using a Grass stimulator which delivered square-wave pulses of 5 msec in duration at a frequency of 16 pulses and an intensity of 4 V. Bipolar electrodes were placed around the auriculotemporal nerve as previously described[7]. Parotid saliva was collected by micropipette directly from the cut end of the duct. Saliva samples were continously collected for the period of 20 min stimula-

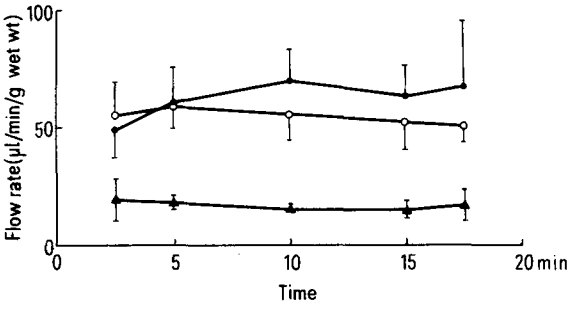

Figure 1. Change in flow rate of parotid saliva evoked by parasympathetic nerve stimulation following chronic administration of amitriptyline. ○—○, control; ●—●, amitriptyline 2 weeks; ▲—▲, amitriptyline 4 weeks.

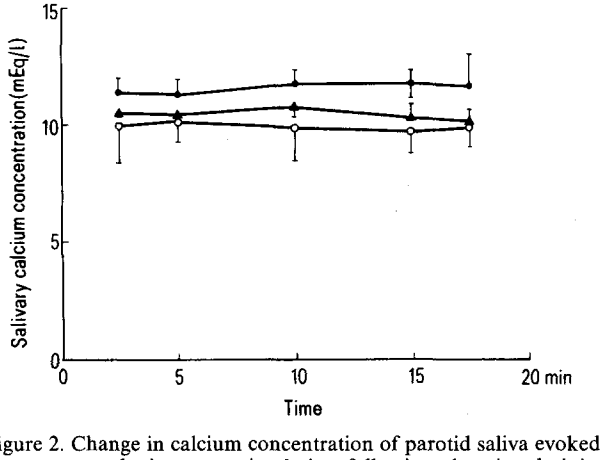

Figure 2. Change in calcium concentration of parotid saliva evoked by parasympathetic nerve stimulation following chronic administration of amitriptyline. ○—○, control; ●—●, amitriptyline 2 weeks; ▲—▲, amitriptyline 4 weeks.

tion. Saliva samples were collected by disposable micropipettes for a timed interval. For the determination of salivary calcium, 10 µl of saliva were used and titrated with ethylene glycol-bis(aminoethyl)-tetraacetic acid (EGTA) with the use of an automatic calcium titrator (Precision Systems). For the analysis of glandular calcium, immediately after 20 min of continuous stimulation of parasympathetic innervation, the stimulated parotid gland was removed rapidly and placed in a crucible and dry-ashed (550 °C for 18 h). The ash was dissolved in 0.5 ml of 1 N HCl and thoroughly mixed. This solution was used for determination of calcium concentration by the automatic calcium titrator. The unstimulated parotid gland of the contralateral side was used as a control and was treated in exactly the same way as the experimental gland.

Results. Data in figure 1 show that there were significant decreases in flow rate of saliva evoked by electrical stimulation of the parasympathetic innervation to the parotid gland of rats treated with amitriptyline (10 mg/kg i.p.) daily for 4 weeks as compared to those of untreated rats (fig. 1). However, parotids of rats treated with amitriptyline for 2 weeks produced a salivary flow in response to such nerve stimulation that was similar to that from glands of untreated rats (fig. 1). Chronic treatment with amitriptyline for either 2 weeks or 4 weeks had no effect on calcium concentration of saliva evoked by parasympathetic nerve stimulation (fig. 2). Chronic amitriptyline treatment did not cause any change in glandular calcium concentration of either control or stimulated parotid (table).

Discussion. The present study shows that chronic treatment with amitriptyline for 4 weeks significantly decreased salivary flow of parotid gland in response to electrical stimulation of the parasympathetic innervation without altering calcium concentration of nerve-evoked saliva. It has been established that there is a parallel decrease in flow rate and calcium concentration of saliva when intensity of stimulation to the parasympathetic innervation to parotid gland is decreased[6]. This presumably is the result of reduction in acetylcholine release. The dissociation between effects on flow rate and calcium concentration suggests that chronic amitriptyline administration may not necessarily be acting by this mechanism. An alternative explanation of these effects is that amitriptyline binds at the same membrane receptors normally occupied by acetylcholine.

Effect of chronic administration of amitriptyline on calcium concentration of rat parotid gland after parasympathetic nerve stimulation

Kind of stimulation	Glandular [Ca] (mEq/kg wet wt) Untreated	Chronic amitriptyline
None	11.80± 0.8 (5)	11.95± 0.2 (4)
Parasympathetic nerve	12.30± 1.0 (5)	11.60± 0.4 (4)

Values are means ± SE. Numbers in parentheses is number of experiments. Parasympathetic innervation to rat parotid (auriculotemporal nerve) was stimulated for 20 min.

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Circadian rhythms of serotonin and the electrical activity of the frontal ganglion of the cockroach, *Periplaneta americana*

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Summary. Rhythmic circadian variations in the spontaneous electrical activity of the frontal ganglion (FG) of the cockroach, *Periplaneta americana*, have been shown, and the neurotransmitter (NT) involved in this activity has been identified as serotonin (5-hydroxytryptamine, 5-HT). During the 24-h day, the diurnal variations in the electrical activity and the levels of 5-HT and its immediate metabolite 5-hydroxyindole acetic acid (5-HIAA) were maximal at 24.00 h and minimal at 12.00 h.

Circadian rhythms in activity are known to play a significant role in insect life. In the cockroach, diurnal rhythms in locomotor activity[1,2] and in spontaneous electrical activity and, in the acetylcholine content in nerve cord[3] have been reported. The FG is a part of the stomatogastric (SG) nervous system of the cockroach and has direct connections with the brain through 2 anterior branches and with the neuroendocrine (NE) system through a posterior recurrent branch. Since it is known that the FG is an autoactive tissue and it fires spontaneously without any signal input[4], its significance for the insect has yet to be established.

Materials and methods. Adult male cockroaches were used for the experiments. They were housed in a cage maintained at a temperature of 25 ± 1 °C in the laboratory. Food and water were supplied regularly. The insects were acclimated to laboratory conditions of 12 h light (08.00–20.00 h) and 12 h dark (20.00–08.00 h) for about 10 days before use in the experiments. The FG was dissected out and kept immersed in cockroach Ringer's solution[5]. Soft sodium glass capillary suction electrodes were used for activity recording whereby the electrical activity of the 2 anterior branches (diameter 100 µm) and that of the posterior recurrent branch (diameter 70 µm) was recorded separately using electrodes of similar diameter. The electrodes were filled with cockroach Ringer's solution and the anterior or the recurrent branch was sucked into the electrode tightly for the recording. To cover the 24-h period, 6 different times (04.00 h, 08.00 h, 12.00 h, 16.00 h, 20.00 h and