etic stimulation were constrictive. In all experiments (25), the greater the stimulation frequency (in range from 6–50 cps), the greater the magnitude of resistance vessel reactions in this vascular zone (Figure 3, r). Capacitance vessel reactions were observed through the whole range of frequencies used. The maximal value of these vessel reactions took place at the frequency of 20 cps, further increase of frequency led to a lesser magnitude of vascular reactions (Figure 3, c). Response latency for resistance vessels amounted to 2.2 ± 0.5 sec and 5.0 ± 0.6 sec for capacitance.

The great individual variability of resistance and capacitance vessel reactions in different animal subjects at the same parameters of electrical stimulation should be noted, too. Thus under stimulation of lumbar sympathetic nerves with the frequency of 15 cps, the perfusion pressure increased by 6 mm Hg with one animal and by 66 mm Hg with another, venous output at the same frequency was 1.4 ml in one case and 5.6 ml in the other.

Results of experiments showed that there is a great difference between frequencies of stimulation, inducing maximal reactions of resistance and capacitance vessels for various vascular zones.

Conclusion. (1) The maximal reactions of cerebral resistance vessels arise at the stimulation frequency of 30 cps, the maximal reactions of capacitance vessels at the frequency of 10 cps. (2) Reactions of resistance vessels in the pulmonary lobe increase in the range of stimulation frequencies from 11–50 cps, maximal reactions of pulmonary capacitance vessels are observed at the frequency of 30–40 cps. (3) Reactions of resistance vessels situated below the abdominal aorta bifurcation increase progressively in the range of frequencies from 6–50 cps, the magnitude of capacitance vessel reaction reaching its maximum value at 20 cps.

Выводы. Максимальные реакции резистивных сосудов мозга возникают при частоте стимуляции 30 имп/сек, емкостных - 10 имп/сек. Реакции резистивных сосудов легкого и расположенных ниже бифуркации брюшной аорты увеличиваются при нарастании частоты стимуляции до 50 имп/сек. Максимальные реакции емкостных сосудов легкого наблюдались при частоте 30-40 имп/сек, сосудов расположенных ниже бифуркации аорты при 20 имп/сек.

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Influence of some Thiamine Antagonists on Frog Taste Receptors

According to VON MURALT and ZOTTERMAN[1], a crude extract of carp intestine containing a thiaminase, strongly inhibits the response of taste receptors to chemical stimuli when applied to the surface of the frog tongue. As a result thiamine (T) has been held to be directly involved in receptor activity.

In order to obtain more conclusive evidence for this hypothesis, we have investigated the influence on taste receptors of other antagonists of T like pyrithiamine (PT) and oxythiamine (OT).

The advantage of PT and OT over thiaminase preparations is that the former are pure compounds whilst the latter may contain several interferring substances.

In a first set of experiments carried out on isolated frog tongue preparations, PT, either applied to the tongue surface or infused intra-arterially, produced only a slight and transient decrease of receptor response to chemical stimuli[2]. However, the isolated frog tongue is not an appropriate preparation for testing the action of competitive inhibitors like PT and OT, because normal excitability of the taste receptors is maintained only for 15–20 min after interruption of the blood supply.

Better experimental conditions were obtained with frog tongue preparations intra-arterially perfused with oxygenated tyrode solution.

Under these experimental conditions, the receptors maintain their response to standard chemical stimuli unchanged over a period of several hours. Furthermore, by this technique it is possible to add the thiamine antagonists in known concentrations to the perfusion medium and to assure their intimate contact with the receptors for the desired length of time.

All the frog's tongue preparations were perfused through the lingual artery for 30 min with amphibian tyrode solutions, so that they might achieve equilibrium under the new conditions.

After 30 min either PT or OT was added to the perfusing medium. Receptor response was tested every 15 min by applying a solution of $CaCl_2$ containing $0.75 \times 10^{-4} M$/l Ca^{++} to the surface of the tongue for 30 sec.

The action potentials were picked up from one of the glossopharyngeal nerves with a suitable pipette, monitored on a Tektronix cathode-ray oscilloscope and counted on an electronic scaler. The perfusion technique and the recording assembly used in these experiments have been described elsewhere[3,4].

Four groups of experiments were carried out to assay the effect of PT on taste receptors. The first group of tongue preparations was perfused with plain tyrode solution. In the other 3 groups, PT, T and PT + T respectively were added to the perfusing medium. The concentration of each active compound in the medium was always $1.2 \times 10^{-4} M$.

Three separate groups of experiments were carried out to study the effect of OT on frog tongue preparations. In 1 set of experiments OT was perfused at a concentration of $2.6 \times 10^{-4} M$, in the other the concentration was higher, $5.3 \times 10^{-4} M$. The OT response was compared with a separate group of control preparations. For each treatment 8 experiments were carried out.

In Figure 1 the activity of the taste receptors, recorded during perfusion in our experiments, is reported as a percentage of the average receptor response obtained for

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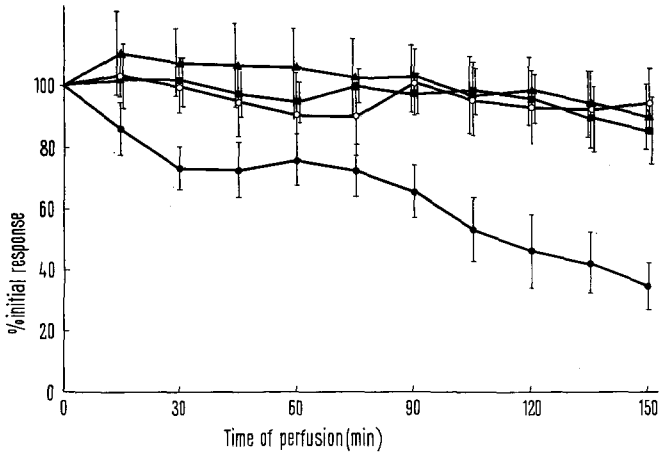

Fig. 1. Response of taste receptors to chemical stimulation (Ca^{++} $0.75 \times 10^{-4} M$) in frog tongue preparations perfused with tyrode solution (○), pyrithiamine $1.2 \times 10^{-4} M$ (●), thiamine $1.2 \times 10^{-4} M$ (▲) and pyrithiamine $1.2 \times 10^{-4} M$ + thiamine $1.2 \times 10^{-4} M$ (■). Standard errors of the means are indicated.

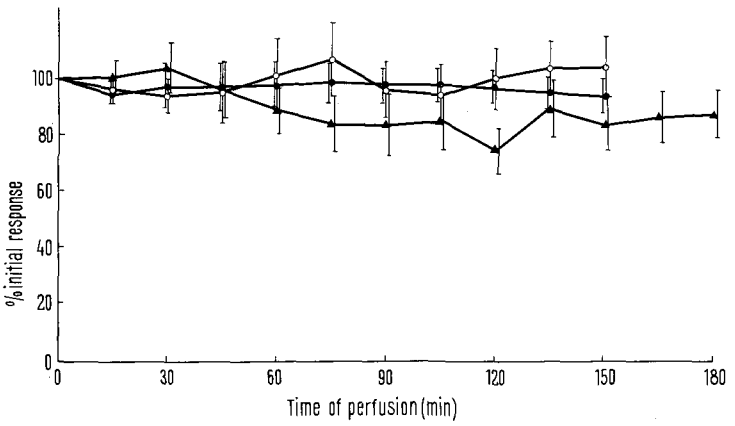

Fig. 2. Response of taste receptors to chemical stimulation (Ca^{++} $0.75 \times 10^{-4} M$) in frog tongue preparations perfused with tyrode solution (○), oxythiamine $2.6 \times 10^{-4} M$ (●), oxythiamine $5.3 \times 10^{-4} M$ (▲). Standard errors of the means are indicated.

each preparation during the initial equilibration period. It can be seen from the data that perfusion with tyrode alone or with $1.2 \times 10^{-4} M$ solution of T did not produce any effect on receptor activity, which was maintained at values near 90% of the initial discharge rate for 2.5 h.

PT at $1.2 \times 10^{-4} M$, on the contrary, produced a reduction of receptor activity, which at the end of the experimental period was down to 34.9% of the starting values.

The fact that there was no such reduction when PT + T $1.2 \times 10^{-4} M$ were perfused demonstrates that the effect is specific. Statistical treatment of the values of receptor discharge shows that the responses to chemical stimuli plotted against time are well represented by a set of straight lines. The lines calculated from the data after tyrode, T, PT + T perfusion have regression coefficients not significantly different from zero. The line obtained with PT perfusion alone has a regression coefficient of -0.4218 with $p < 0.001\%$.

OT treatment at both the concentrations used (Figure 2) produced no significant reduction of receptor discharge as compared with its own controls.

Our results are consistent with the hypothesis that thiamine is an essential factor in the process of taste receptor excitation. This is clearly proved by the specific inhibition exerted by PT on their response to chemical stimuli.

The lack of any depressing action of OT can be explained by considering that OT acts as a competitive antagonist of cocarboxylase only when phosphorylated and that its rate of in vivo phosphorylation is very low[5]. On the other hand, it is well known that in vivo PT actively inhibits the phosphorylation of T by the thiaminokinase system[6].

Both the depressing action of pyrithiamine and the lack of any influence of oxythiamine suggest that thiamine is involved in the excitation processes of taste receptors only in the phosphorylated form.

Riassunto. La piritiamina su preparazioni di lingua di rana perfuse deprime notevolmente la risposta dei recettori gustativi alla stimolazione chimica. Questo effetto è dovuto alla sua azione antitiaminica perchè scompare in presenza di quantità equimolecolari di tiamina. L'ossitiamina al contrario non ha mostrato questo azione.

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