Pharmacological Analysis of Pentagastrin-Modulated Behavior Caused by Stimulation of the Ventromedial Hypothalamus

V. G. Zilov and A. P. Patyshakuliev

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Key Words: *pentagastrin; avoidance response; blockers of transmitters; rabbits*

Some oligopeptides are known to alter temporarily and selectively the nature of behavioral responses caused by electrical stimulation of different motivation centers of the hypothalamus [4].

In the present work we attempted to study the ability of pentagastrin (PG) to alter the avoidance response for threshold electrical stimulation of the ventromedial hypothalamus (VMH). PG localized in the region of the hypothalamus [8] and in other brain structures [15] which play an important role in the genesis of biological motivations is able to enhance food consumption in animals [6]. Thus, in the present experiments the excitation underlying the defense motivation for stimulation of the VMH encounters, as it were, the "food" excitation arising in response to PG administration. Of interest is not only the result of such an interaction, which must be reflected in the pattern of behavioral response evoked in the hypothalamus, but also an analysis of the possible participation of certain classic transmitters of the central nervous system (CNS) in the induced behavior.

MATERIALS AND METHODS

The experiments were carried out on 17 male rabbits weighing 2.5-3 kg under conditions of free behavior. Preliminarily fed animals were used in the experiment. Bipolar Nichrome electrodes 0.1 mm in diameter were implanted according to the rabbit brain atlas (K. N. Soyer *et al.*, 1954) in the ventromedial region of the hypothalamus of a preliminarily scalped animal. The threshold values of stimulation of the VMH which were required to induce the avoidance response in the animals varied in the range of 1.5-3.5 V (frequency 50 Hz, pulse duration 1 msec) depending on the accuracy of implantation. A single injection of PG (Serva, Germany) in a dose of 35 μg/kg in 20 μl of saline was performed with the aid of a microsyringe via a cannula implanted in the lateral ventricle of the brain. In the control series of experiments, physiological saline (20 μl) was injected in the lateral ventricle in order to reveal the specificity of the effect of PG. In the present experiments the ampulated solutions of inderal (Imperial Chemical Ind., Ltd., Great Britain), calypsol (Gedeon Richter, Hungary), atropine (Russia), as well as baclofen (Polfa, Poland) and ketanserin (Jansser, Denmark) were used. The preparations were administered in doses of 0.25 and 0.5 mg/kg (inderal, calypsol, and baclofen), 0.5 and 1 mg/kg (atropine), and 0.1 mg/kg (ketanserin) intravenously. The excitability of the VMH and the latencies of the behavioral responses evoked there were assessed at the end of intraventricular injection of PG and every 5 min over a 2.5-h period after injection of the antagonists. The experimen-

Department of Normal Physiology, I. M. Sechenov Moscow Medical Academy. (Presented by K. V. Sudakov, Member of the Russian Academy of Medical Sciences)

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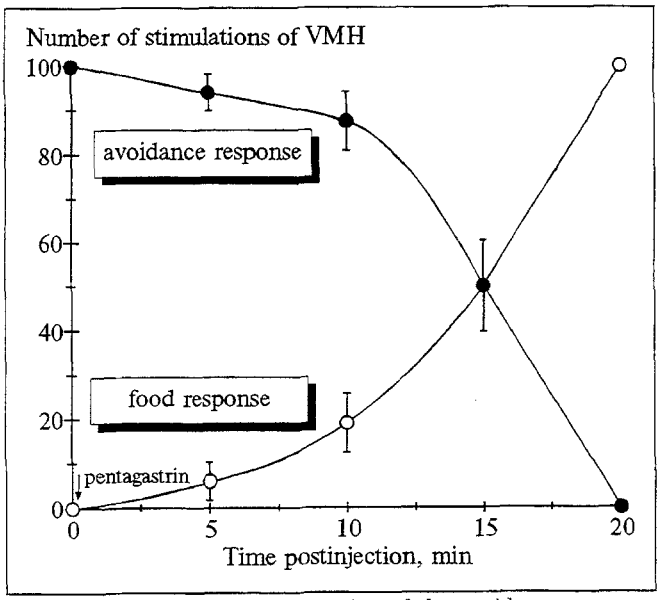

Fig. 1. Dynamics of transformation of the avoidance response into a food response for stimulation of the VMH against the background of single intraventricular injection of PG in a dose of 35 μg/kg.

tal results were mathematically processed by statistical analysis on a PC. The localization of the subcortical electrodes was determined using the express method, by preparing sections of the brain 50-100 μ thick.

RESULTS

In the animals electrical stimulation of the VMH caused an avoidance response (AR) after a short-term orienting-exploratory response. The administration of PG to preliminarily fed animals transformed the AR induced by the threshold electrical stimulation of the VMH into a food response ($p<0.01$).

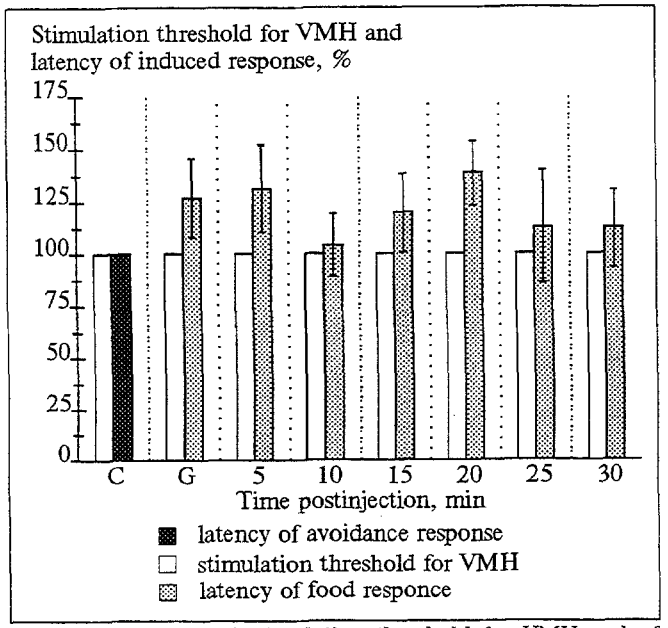

Fig. 2. Dynamics of stimulation threshold for VMH and of latency of AR and food response after a single injection of PG in a dose of 35 μg/kg and after intravenous injection of 0.5 mg/kg inderal.

In the present study we analyzed the time it took for the AR to be transformed into a food response, after a single injection of PG into the brain ventricle, which turned out to be 20 min ($p<0.05$). The dynamics of the decrease of occurrence of AR and, conversely, of the increase of occurrence of the food response during stimulation of the VMH is presented in Fig. 1.

Like other oligopeptides [4] PG exhibited a reversible ability to transform the AR into a food response, which lasted altogether for 150-155 min after the end of intraventricular injection ($p<0.05$).

The experiments in which the substances exhibiting properties of β-adrenoblockers (inderal) and the M-cholinoblocker atropine were used enabled us to establish that neither β-adrenergic nor M-cholinergic mechanisms are crucial in the PG-mediated transformation of the AR into a food response ($p<0.001$, Fig. 2).

A different result was obtained for injection of the serotonin antagonist ketanserin and of calypsol (ketamin), a substance with a complex mechanism of action but with pronounced effects of an N-cholinoblocker (Fig. 3). For instance, after intravenous infusions of calypsol in doses of 0.25 and 0.5 mg/kg were finished, electrical stimulation of the VMH once again evoked an AR in the animals, despite the preceding injection of PG ($p<0.001$, Fig. 3). We failed to discover any dose-dependent effect for the use of the above concentrations of calypsol.

The effect of baclofen (one of the GABA antagonists) proved to be specific: this preparation reduced the time of PG-induced transformation but had no effect on the PG-altered pattern of behavioral response. For instance, against the background of baclofen electrical stimulation of the VMH following the injection of PG evoked a food response, but the recovery of the AR for stimulation of the VMH occurred 30 min earlier ($p<0.01$).

Summarized results of the experiments are presented in Table 1. Our findings indicated the ability of one more substance of a peptide nature to alter temporarily and selectively the pattern of behavioral response to stimulation of the VMH in preliminarily fed animals. This assumption is consistent with earlier observations [5] which demonstrated the effects of different neuropeptides on the defense responses caused by electrical stimulation of the VMH. In particular, the ability of PG to

TABLE 1. Effects of Blockers on PG-Induced Transformation Phenomenon

Experimental conditions (n=5)	Blocker	Final response
PG-induced food response for stimulation of VMH	Inderal	food response
	Atropine	– " –
	Baclofen	– " –
	Calypsol	avoidance response
	Ketanserin	avoidance response

affect the defense responses, depending on whether food motivation had been satisfied, was noted in rabbits.

However, the mechanisms of PG transformation of the AR into a food response are still to be elucidated. One possible explanation is that PG interacts with classic transmitters of the CNS. The interaction of PG and epinephrine was established in experiments on individual neurons of the lateral hypothalamus [1], the nature of the modulatory effect of the neuropeptide depending on the initial state of the animal. The ability of cholecystokinin and its C-terminus (PG) to alter the sensitivity of the dopaminergic receptors in the brain was noted [11]. A close relationship between PG and dopamine in the brain is indicated by observations on the coexistence of these substances in the neurons of the mesolimbic system [10]. PG is able to influence the activity of cholinesterase in the cerebral cortex of rats [12]. The level of serotonin in the alimentary tract, blood, and brain is also determined by the concentration of PG [14].

Our studies enabled us to conclude that the β-adrenergic and M-cholinergic structures do not play any important role in the mechanisms of PG-induced transformation of the AR into a food response, whereas calypsol (a substance with the effect of an N-cholinergic antagonist) and ketanserin (a serotonin antagonist) are conducive to the recovery of the AR during electrical stimulation of the VMH, despite the preceding injection of PG.

GABA-ergic structures of the brain probably do not play a crucial role in the PG-mediated transformation of the AR into a food response, but the use of baclofen in the experiments considerably reduced the time of the PG effect.

Today, classical transmitters are regarded as only one of the possible mechanisms of the effect of PG on motivational behavioral responses in animals. In addition, PG interacts with two types of cholecystokinin receptors, the concentration of which is especially high in the cerebral cortex, dentate gyrus, olfactory bulb, thalamus, and hypothalamus [9,13]. It is important to note that B-type cholecystokinin brain receptors are involved in the realization of the responses accompanied by anxiety and negative emotions [16].

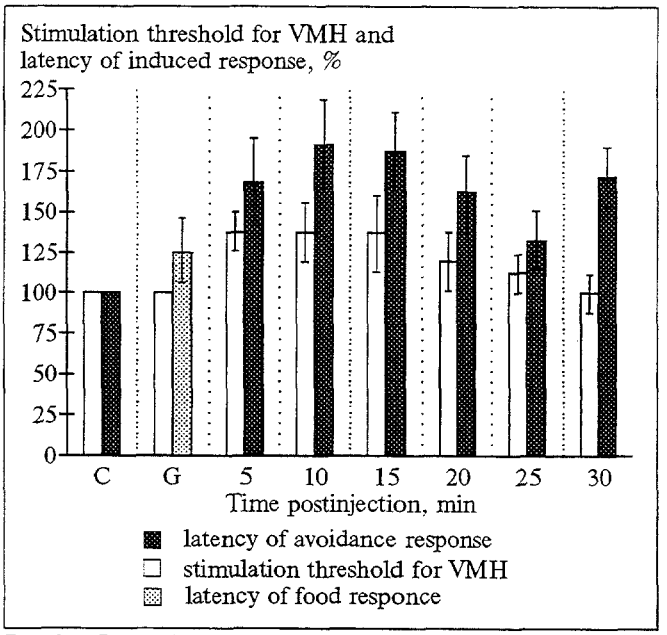

Fig. 3. Dynamics of stimulation threshold of VMH and of latency of AR and food response after a single injection of PG in a dose of 35 μg/kg and intravenous injection of 0.5 mg/kg calypsol.

One must not disregard numerous recently reported examples of the interaction of PG with other oligopeptides in the brain for the implementation of various functions [3]. In particular, the stimulating effect of PG on the production of growth hormone and prolactin [7] is established, as well as the coexistence of gastrinlike peptides with other peptides in the pituitary and hypothalamus [17].

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MICROBIOLOGY AND IMMUNOLOGY

Mechanisms of Immunotropic Effects of Organophosphorus Compounds

P. F. Zabrodskii

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In connection with the wide application of organophosphorus compounds (OPC) in agriculture and the chemical industry and the destruction of OPC classified as toxic substances, with the concomitant risk of acute and chronic intoxications (in particular, during possible accidents), there is unflagging interest in investigations of the mechanisms of action of the above toxicants on the immune system. Such studies are aimed at the development of methods of preventing and treating postintoxication immunodeficient states accompanied by various infectious complications and diseases [5]. However, analysis of recent publications on the immunosuppressive effects of OPC [2,4,11] indicates that much remains unclear regarding the significance of nonspecific (associated with the stress response) and specific mechanisms in the development of disturbances of the immune status during exposure to OPC.

The aim of the present study was to assess the role of nonspecific and specific mechanisms in the formation of the principal immune responses to OPC.

MATERIALS AND METHODS

The experiments were carried out on male CBA mice weighing 18-22 g. The anti-cholinesterase insecticide dimethylchlorvinylphosphate (DDVP) in doses of 0.25, 0.5, and 1.0 of LD_{50} (40±1.2 mg/kg, subcutaneously) was used as the OPC. The immune responses to DDVP were compared with the

Department of Toxicology, Saratov Medical Institute. (Presented by A. D. Ado, Member of the Russian Academy of Medical Sciences)

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