at once that on application of NA to segments L_4-S_1 its action on reflexes evoked by stimulation of TN did not differ in principle from its action on reflexes from RN, examined in detail above, on application to segments C_6-T_1. Comparison of the mean values of PCR for these experiments to identical stimulation of the same nerve (RN or TN) before the action of NA and at the time of maximal development of its effect (Fig. 3) shows that on application of NA to segments C_6-T_1 the magnitude of PCR evoked by stimulation of TN did not change statistically significantly, whereas on application of NA to segments L_4-S_1 PCR to stimulation of RN did not change statistically significantly. Consequently, under these experimental conditions depression of PCR to volleys of spinal afferents is in fact due to a local change in the processing of afferent signals.

We do not know how the NA concentration falls as it penetrates into the spinal cord, or what concentration of NA really acts under these experimental conditions. However, it may be noted that release of endogenous NA in the region of the dorsal horns of the spinal cord also leads to depression of PCR induced by volleys of impulses in spinal afferents, as the writers have shown.

LITERATURE CITED

1. Yu. B. Gailans, "Investigation of suprabulbar regulation of vasomotor reflexes evoked by impulses in somatic afferent fibers," Author's Abstract of Dissertation for the Degree of Candidate of Medical Sciences, Moscow (1978).
2. V. M. Khayutin, R. S. Sonina, and E. V. Lukoshkova, Central Organization of Vasomotor Control [in Russian], Moscow (1977).
3. G. Belcher, R. W. Ryall, and R. Schaffner, Brain Res., 151, 307 (1978).
4. P. M. Headley, A. W. Duggan, and B. I. Griersmith, Brain Res., 145, 185 (1978).
5. L. I. Kleinman and E. P. Radford, J. Appl. Physiol., 19, 360 (1964).
6. Y. Kuraishi, Y. Harada, and H. Takagi, Brain Res., 174, 333 (1979).
7. E. V. Lukoshkova and G. Pavlic, Pflüg. Arch., 336, 134 (1972).
8. S. V. R. Reddy and T. L. Yaksh, Brain Res., 189, 391 (1980).
9. H. Weber, Verh. Dtsch. Gesell. inn. Med., 21, 616 (1904).
10. W. D. Willis, Control of Nociceptive Transmission in the Spinal Cord, Berlin (1982).

EFFECT OF DELTA-SLEEP PEPTIDE ON PARASYMPATHETIC REGULATION OF THE HEART RATE

M. A. Zvyagintseva, I. L. Kosharskaya, and L. S. Ul'yaninskii

UDC 612.178.3.014.46:615.31.547.96

KEY WORDS: delta-sleep peptide; extracardial regulation

To restore normal sleep in patients with various sleep disturbances, delta-sleep peptide (DSP) has begun to be used in recent years [7, 8]. Meanwhile, as experimental studies of this peptide have shown, it has a marked prophylactic action in negative emotional states [1, 4]. DSP can therefore be used in clinical practice for the prevention and treatment of stress. However, its effect on various functional systems and, in particular, on the cardiovascular system is not yet clear, although there is information to show that DSP can slow the heart and respiration rates and reduce oxygen consumption in unrestrained animals [2].

The aim of this investigation was to study the effect of DSP on parasympathetic regulation of cardiac activity.

Laboratory of Experimental Cardiology, P. K. Anokhin Research Institute of Normal Physiology, Academy of Medical Sciences of the USSR, Moscow.

(Presented by Academician of the Academy of Medical Sciences of the USSR G. N. Kryzhanovskii.) Translated from Byulleten' Eksperimental'noi Biologii i Meditsiny, Vol. 101, No. 4, pp. 390-391, April, 1986. Original article submitted March 15, 1985.

0007-4888/86/0004-0402$12.50

EXPERIMENTAL METHOD

Experiments were carried out on 28 chinchilla rabbits. Changes in the cardiac rhythm under the influence of DSP were studied in 6 unrestrained rabbits in a quiet state. Acute experiments were conducted on 22 animals anesthetized with pentobarbital. Of this number, 6 rabbits received the peptide during the action of the anesthetic, and changes in heart rate (HR) arising under these circumstances were recorded. In the remaining 16 rabbits the action of DSP on the effects of vagus nerve stimulation was studied. The peripheral end of the divided left or right vagus nerve was stimulated for 30 sec. The effect of vagal inhibition was estimated from the duration of the pause or the degree of slowing of HR after electrical stimulation of the vagus nerve for 10, 20, and 30 sec, respectively. The vagus nerve was stimulated electrically by pulses with a frequency of 100 Hz and duration 2-3 msec; the strength of the current was chosen to be a little above threshold for inducing a definite vagus effect. Effects of vagus nerve stimulation were analyzed before and after injection of the DSP, every 10 min for 1.5-2 h. DSP was injected intravenously in a dose of 60 nanomoles/kg [1, 3]. The ECG in standard lead II, the aortic pressure, the left ventricular pressure (LVP) and its first derivative, the respiration rate, and the strength of the current used to stimulate the vagus nerve were all recorded on a Mingograf-82.

EXPERIMENTAL RESULTS

Injection of DSP into 6 rabbits in a dose of 60 nanomoles/mg when the animals were unrestrained and in a quiet state caused a moderate (by 16%) fall of HR from 228 ± 32.4 to 190 ± 26.4 beats/min. This slowing of the heart was observed at the 10-20th minute of action of the peptide and it was abolished by injection of atropine.

In acute experiments on 6 rabbits under anesthesia HR rose from 228 ± 32.4 to 291 ± 22.0 beats/min. Injection of DSP against this background also caused a moderate (by 12%) fall of HR from 291 ± 22.0 to 255 ± 36.9 beats/min. This effect was abolished by injection of atropine or by division of the vagus nerve.

The action of DSP on the effects of stimulation of the vagus nerves was investigated in acute experiments on 16 rabbits. After bilateral division of the vagus nerves HR rose to 312 ± 14.5 beats/min. Under these conditions it fell by 8% under the influence of DSP, and 60 min after injection of the peptide it was 287 ± 13.9 beats/min. The intraventricular and aortic pressures also fell somewhat, whereas the respiration rate remained at a relatively stable level.

Injection of DSP had a marked effect on vagal inhibition of cardiac activity: during the action of the peptide lengthening of vagus arrest of the heart or an increase in the degree of the negative chronotropic effect was observed. This action began to appear during the first few minutes after injection of the peptide, and it gradually intensified over a period of 1.5-2 h. At the beginning of action of the peptide, transient fluctuations of the heart rate followed by stable slowing of the heart could be observed. This is illustrated by one

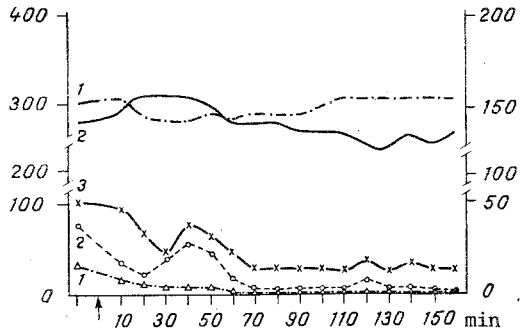

Fig. 1. Strengthening of vagal inhibition of cardiac activity in rabbit under the influence of DSP. Abscissa, time (in min); ordinate: on left, HR (beats/min), on right, LVP (in mm Hg). a) HR (1) and LVP (2) before injection of DSP and during its action; b) effect of stimulation of vagus nerve on HR before injection of DSP and during its action: 1, 2, 3) 10, 20, and 30 sec of stimulation, respectively. Time of injection of DSP indicated by arrow. Duration of each period of stimulation of vagus nerve 30 sec, frequency of stimulation — every 10 min over a period of 1.5-2.5 h.

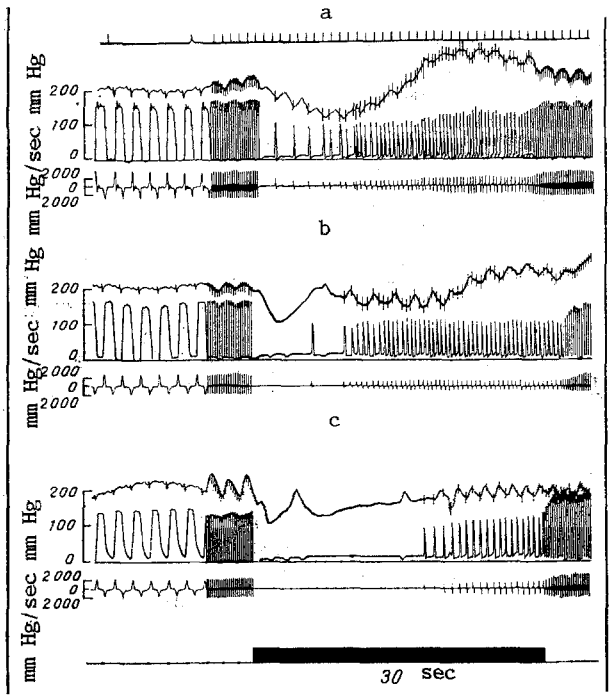

Fig. 2. Development of vagal arrest of the heart in a rabbit under the influence of DSP: a) Effects of stimulation of vagus nerve before injection of DSP; b, c) the same after injection of DSP: slowing of HR during control stimulation of vagus nerve (a); during stimulation of vagus nerve preceded by injection of DSP (60 nanomoles/kg) — ventricular arrest for 7 sec occurred after 10 min (b) and after 1.5 h duration of arrest increased to 20 sec (c). For all fragments: above — time marker, 1 sec; below — marker of stimulation of vagus nerve. For each fragment, from top to bottom: ECG, pressure in left ventricle (in mm Hg), and its first derivative (in mm Hg/sec).

experiment in which, during control stimulation of the vagus nerve, HR for 10 sec was 36 beats/min, for 20 sec 75 beats/min, and for 30 sec 100 beats/min. Under the influence of DSP, HR fell after 10 min to 18, 57, and 92 beat/min for 10, 20, and 30 sec of stimulation, respectively. During subsequent stimulation of the vagus nerve HR became slower still, and 90 min after injection of the peptide it was 0, 3 , and 26 beats/min for 10, 20 and 30 sec, respectively (Figs. 1 and 2).

This marked action of DSP on vagal inhibition was observed in 14 of 16 experiments. In two experiments the peptide caused quickening, not slowing, of the heart. The effect observed was independent of which vagus nerve — right or left — was stimulated.

DSP thus has both a central and a peripheral action on parasympathetic regulation of the heart, strengthening vagal inhibition of the cardiac rhythm. Slowing of the heart under the influence of DSP has been observed by other workers also, after direct injection of the peptide into the cerebral ventricles [2]. When administered this way, the negative chronotropic effect of the peptide was stronger, and consequently, the intensity of the effect of the peptide depends on the method of its administration.

The moderate enhancement of parasympathetic regulation under the influence of DSP may be a favorable factor in emotional stress. As the writers showed previously [5, 6], moderate predominance of parasympathetic influences during stress leads to elevation of the thresholds of onset of cardiac arrhythmias and also prevents the increase in catecholamine concentration and the development of structural disturbances in the myocardium.

LITERAUTRE CITED

1. E. V. Koplik, D. F. Vedyaev, I. I. Mikhaleva, et al., Dokl. Akad. Nauk SSSR, 267, 230 (1982).

2. V. L. Medvedev and V. D. Bakharev, Zh. Evol. Biokhim. Fiziol., 15, 379 (1979).
3. A. S. Sargsyan, L. V. Sumskaya, and I. Yu. Aleksandrova, Bioorg. Khim., 7, 1125 (1981).
4. K. V. Sudakov, V. T. Ivanov, V. I. Badikov, et al., in: Stress, Adaptation, and Functional Disturbances [in Russian], Kishinev (1984), p. 357.
5. L. S. Ul'yaninskii, E. Kellerova, I. L. Kosharskaya, et al., in: Stress, Adaptation, and Functional Disturbances [in Russian], Kishinev (1984), p. 357.
6. L. S. Ul'yaninskii (L. S. Ulyaninsky), N. N. Beskrovnova, V. A. Kuznetsova, et al., in: Sudden Cardiac Death. Third USA–USSR Joint Symposium, Kaunas (1982), p. 361.
7. D. Schneider-Helmert, F. Gnirrs, M. Monnier, et al., Int. J. Clin. Pharmacol., 19, 341 (1981).
8. D. Schneider-Helmert and G. Schoenenberger, Neuropsychobiology, 9, 197 (1983).

IMMUNOHISTOCHEMICAL IDENTIFICATION OF ENDOGENOUS SOURCES OF CARDIAC GLYCOSIDE BIOSYNTHESIS

I. M. Kvetnoi, I. P. Kololyuk, and G. M. Deineko

UDC 612.822.015.36:547.918]-088.1

KEY WORDS: cardiac glycosides; immunohistochemical identification; endogenous sources.

As a result of progress in the development of immunohistochemical methods of investigation it is now possible to identify sources of biosynthesis and storage of chemical substances with antigenic properties, including hormones, proteins, enzymes, and so on, in man and animals. The data thus obtained have considerably widened our ideas on pathways of synthesis and metabolism of many biologically important products, and compelled a re-examination of some existing views, and some new and hitherto unknown compounds have been discovered [1, 5, 6].

This paper describes an attempt to discover endogenous sources of synthesis of substances with physiological and pharmacological properties characteristic of cardiac glycosides. The theoretical grounds for such a search were provided by data on synthesis of endorphins – substances pharmacologically similar to opiates of plant origin, in animals and man [3, 4].

EXPERIMENTAL METHOD

Material for investigation was taken from various parts of the brain (hypothalamus, cerebellum, brain stem, medulla), the atrial wall, the superior vena cava where it enters into the right atrium (the zone of the sinoatrial node), the lungs, liver, pancreas, stomach, different parts of the intestine, and the adrenals and kidneys of dogs. Pieces of the above-mentioned organs were fixed in 10% neutral formalin and Bouin's fluid and embedded in paraffin wax. Dewaxed sections were stained with hematoxylin and eosin and by the argyrophilic method of Grimelius. Sections cut from material fixed in Bouin's fluid were treated immunohistochemically with the use of antiserum against digoxin from the DIGOCTK-125 kit (from CIS, France). To confirm the specificity of the immunohistochemical reaction, the usual controls were set up, including exhaustion of the antiserum with a pharmacopoeial solution of digoxin. Luminescent donkey serum against rabbit globulins, labeled with fluorescein isothiocyanate, was used as the label. The preparations were examined in the LYUMAM I-3 luminescent microscope (LOMO, USSR) with FS 1-2 and SZS 7-2 filters.

Research Institute of Medical Radiology, Academy of Medical Sciences of the USSR, Obninsk. D. I. Ul'yanov Kuibyshev Medical Institute. (Presented by Academician of the Academy of Medical Sciences of the USSR I. B. Soldatov.) Translated from Byulleten' Eksperimental'noi Biologii i Meditsiny, Vol. 101, No. 4, pp. 392-393, April, 1986. Original article submitted February 11, 1985.

0007-4888/86/0004-0405$12.50