The results show that a decrease in the amplitude of the evoked potential as a result of collision takes place not only in the region between the electrodes (L_1). A decrease in amplitude can also be observed when the colliding impulse has already passed the stimulating electrodes for A_β-fibers by 71.7 mm and for A_δ-fibers by 143.1 mm, and for C-fibers by 44.4 mm. This fact is very important for the colliding impulses method, for it increases the probability of collisions between antidromic and orthodromic impulses and increases the distances allowable between stimulating and recording electrodes. In this way the limitations on the use of the colliding impulses method can be reduced. The previous theoretical calculations [1, 3] did not take into account the duration of the refractory state of impulses spreading along the nerve, and considered the impulse to be single point.

Thus model experiments and calculations based on them have widened the scope for use of the method by reducing the limitations imposed by the length of the interelectrode segment and of the whole nerve.

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DISTRIBUTION OF LABELED AMINO ACIDS AND DELTA SLEEP-INDUCING PEPTIDE IN THE BODY AFTER INSTILLATION INTO THE RABBIT CONJUNCTIVA

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Our previous investigations [1, 3] showed that instillation of the regulatory peptides angiotensin-II and delta sleep-inducing peptide (DSIP) into the conjunctival sac of rabbits gives rise to physiological effects similar to those observed when they are administered by the intragastric and intravenous routes. These observations were confirmed by other investigators [2], but the problem of the pathways of spread of oligopeptides in the body and their effects on the various physiological functions when applied to the conjunctiva of the eye remained unsolved.

The aim of this investigation was to study the distribution of individual amino acids and of DSIP, labeled with tritium, when instilled into the conjunctival sac of the rabbit's eye.

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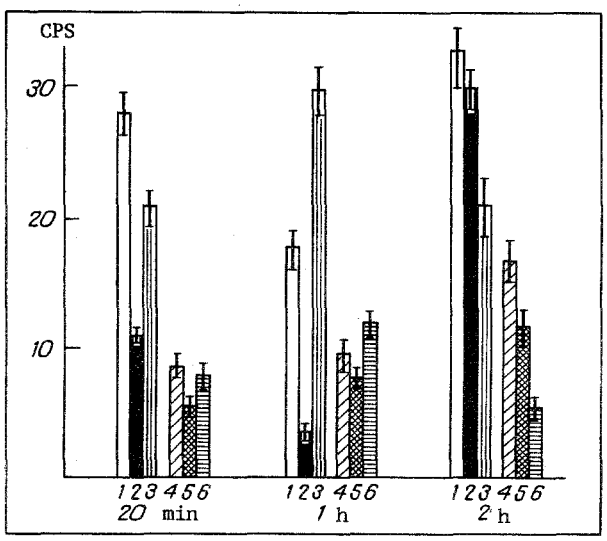

Fig. 1. Distribution of labeled amino acids in various brain structures depending on time after their instillation into conjunctival sac (n = 10). 1) Activity of ^{3}H-valine in visual cortex, 2) in thalamus, 3) in optic chiasma, 4) activity of ^{3}H-glycine in visual cortex, 5) in thalamus, 6) in optic chiasma.

EXPERIMENTAL METIIOD

Experiments were carried out on 18 adult chinchilla rabbits weighing 3 kg. A solution of ^{3}H-valine, ^{3}H-glycine, or ^{3}H-DSIP in distilled water, with total activity of 1 mCi in a volume of 0.1 ml, was instilled bilaterally into the conjunctival sac of rabbits lightly restrained in a frame. At intervals of 10, 20, and 30 min and 1, 2, and 6 h after administration of these substances, CSF was removed from the animals intravitally in a volume of 0.5 ml from the lateral ventricles and 1 ml of blood was taken from the marginal vein of the ear. The animals were decapitated, and when the same times had elapsed after administration of the substances, pieces of the brain (visual cortex, thalamus, optic chiasma), heart (left ventricle), and spleen (right or left quadrant) were taken for analysis. The structures to be studied were homogenized in buffered physiological saline (pH 7.2). Peptides were extracted with 4 volumes of an ethanol:acetone (1:1) mixture. Activity of the supernatant after centrifugation was determined in toluene scintillation solution by means of a 1215 scintillation spectrometer (LKB, Sweden). The method of analysis of the CSF and serum was identical.

EXPERIMENTAL RESULTS

The experiments showed that both when 0.1 ml of a solution of ^{3}H-valine and ^{3}H-DSIP were instilled into the conjunctival sac, after 10 min they were found to be present in all media and tissues tested. The intensity of radioactivity in the different tissues differed, however, depending on the time elapsing after instillation.

The maximal concentration of ^{3}H-valine was found in the visual cortex and in the thalamus toward the end of the 2nd hour after instillation. The peak of radioactivity in the optic chiasma was observed toward the end of the 1st hour, and this was followed by a decrease in the 2nd hour of observation. A similar distribution of activity among the various brain structures was found for ^{3}H-glycine (Fig. 1).

The maximal blood and CSF levels of ^{3}H-DSIP were recorded after 20 min, and the minimal levels 2 h after its instillation into the conjunctival sac.

The distribution of ^{3}H-DSIP in various structures of the brain, heart, and spleen depending on the time elapsing after instillation into the conjunctival sac is shown in Fig. 2. The maximal intensity of radioactivity in the tissues of the heart, spleen, and optic chiasma was recorded in the first 30 min. The peak of radioactivity in the thalamus was observed 2 h after injection of ^{3}H-DSIP. Radioactivity in tissues of the visual cortex increased toward the end of the 1st hour and gradually reached a maximal by the 6th hour of observation.

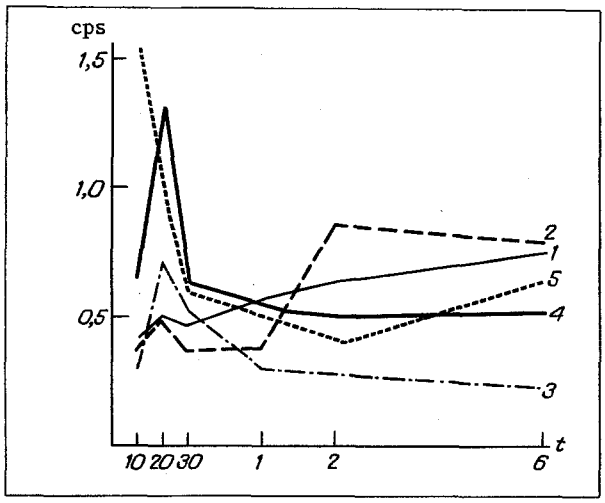

Fig. 2. Distribution of ^{3}H-DSIP in various brain structures and tissues of the heart and spleen depending on time elapsing after instillation into conjunctival sac (n = 8).

The rapid rise of radioactivity in the CSF and blood and also in tissues of the heart, spleen, and optic chiasma may be evidence of penetration of the isotope from the conjunctival sac directly into the bloodstream, followed by its distribution among the brain structures. The possibility of penetration of DSIP through the blood-brain barrier has been demonstrated radioimmunologically [4]. This does not rule out other possible ways of spreading of DSIP directly into the brain on account of diffusion along nerve fibers, as has been demonstrated for ^{3}H-fructose and 2,3-^{3}H-proline when injected directly into the vitreous body [5].

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