Short Communication

Studies on the Effect of Glutamic, Aspartic and γ-Amino Butyric Acids on Locust Muscle Fibre Membrane Potential

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We have studied the effect of various concentrations of L-glutamic (GLU), L-aspartic (ASP), γ-amino butyric acids (GABA), and L-aspargine (ASN) and L-glutamine (GLN) on the membrane potential (V_m) of muscle fibres dissected from three different muscles in the jumping leg of *Locusta migratoria asiatica*: m.flexor tibiae, m.extensor tibiae and m. retractor unguis.

Dissection of muscle fibres and measuring of their electric parameters were performed according to Zachar et al. (1978) at temperatures of 24 ± 1°C. The basic

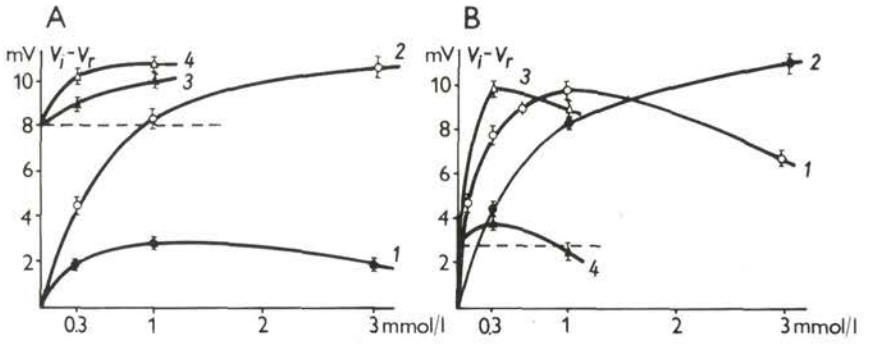

Fig. 1. Effects of GLU, ASP and GABA on the membrane potential of the flexor tibiae muscle fibre. *Abscissa*: concentration of the tested substances (mmol/l). *Ordinate*: membrane depolarization (V_i—V_r) (in mV), where: V_i — is the membrane potential during perfusion with GLU, ASP or GABA; V_r — is the membrane resting potential. Each point represents the mean from 5 — 12 measurements ± SD. V_m was measured with an accuracy of 0.1 mV. *A*:1 — GABA; 2 — ASP; 3,4 — GABA and ASP, respectively, in the presence of 0.3 mmol/l GLU. Interrupted line shows the depolarization level in the presence of 0.3 mmol/l GLU. *B*:1 — ASP; 2 — GLU; 3,4 — GLU and ASP, respectively, in the presence of 1 mmol/l GABA. Interrupted line shows the depolarization level in the presence of 1 mmol/l GABA.

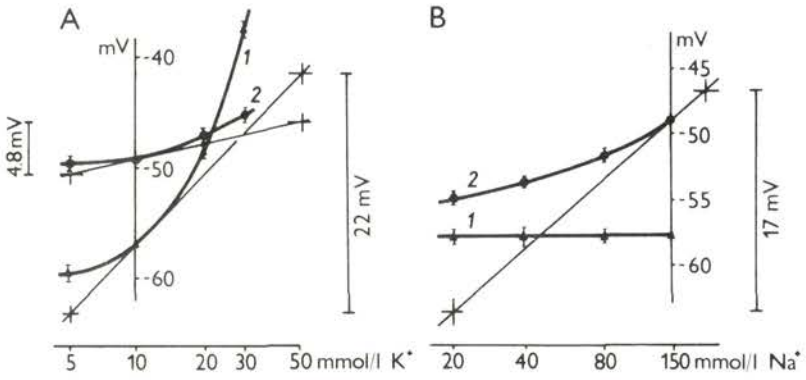

Fig. 2. The dependence of membrane potential of the locust flexor tibiae muscle fibre on the external concentration of K^+ (*A*) and Na^+ (*B*) ions at constant concentrations of Cl^-.
Abscissa: concentration (in mmol/l) of K^+ (*A*) or Na^+ (*B*) ions.
Ordinate: membrane potential (in mV). The slopes to the exponential curves are considered to be indices of the relative membrane conductance for an ion at a sudden change in concentration. Mean ± SD values from 5 — 12 measurements are given. *A*. Dependence of membrane potential on K^+ ion concentrations in physiological solution in the absence (1) or presence of 0.3 mmol/l GLU (2). *B*:Dependence of membrane potential on Na^+ ion concentrations in physiological solution in the absence (1) or presence of 0.3 mmol/l GLU (2).

solution contained (in mmol/l): 10 K^+, 156 Na^+, 152 Cl^-, 10 HPO_4^{2-}, 1.3 Ca^{2+}, pH 6.8 (Usherwood and Machili 1968). Complete exchange of the bathing solution was accomplished within 10 seconds. Fig. 1 illustrates the depolarizing effect of GLU, ASP and GABA in the flexor tibiae muscle fibres. The effect was qualitatively identical in all the three muscles studied. ASN and GLN were, however, ineffective even at concentrations of 10 mmol/l. The depolarizing effect of GLU and ASP was in good agreement with the data reported in literature (Beránek and Miller 1968; Usherwood 1977). There was, however, a discrepancy between the depolarizing effect of GABA and its inhibitory action (Grundfest and Usherwood 1965; Usherwood and Grundfest 1965). It should be noted that the effect of GABA did not change if the recording microelectrode was filled with 3 mol/l of CH_3COOK. The absolute value of the zero current potential of Cl^- ions is likely lower than the resting potential. Voltage (V_m)-dependence upon the combined action of GLU, ASP and GABA were studied in order to establish whether there exist specific receptors. In agreement with Usherwood (1977) we have established that the effects of GLU and ASP are not additive, whereas the effects of relatively low concentrations of GLU and GABA sum, indicating the presence of specific GABA receptors in the membrane. The inhibitory effect of GABA was demonstrated with higher GLU concentrations. This effect was more pronounced during the co-action of ASP and GABA on the GLU-receptor. This might be taken as evidence for the inhibitory action of GABA on locust neuro-muscular transmission despite its depolarizing effect.

Fig. 2 shows the effect of GLU on the relative conductances of the locust muscle fibre membrane for K^+ and Na^+ ions. The procedure as described by Zachar et al. (1978) was used. It was shown that the transport number (T_K) for K^+ ions decreased (from 0.38 to 0.08), while that for Na^+ ions increased (from 0.0 to 0.29) in the presence of GLU. The results are in agreement with those of Tishchenko (1977) and McDonald (1975). The difference between our T_K value for muscle fibres in normal state and the results of Zachar et al. (1978) may be explained by slower exchange of solutions in our experiments.

Two conclusions can be drawn from these results. First, two carboxyl groups in molecules of GLU and ASP are necessary for conversion of the receptor-operated channel into the conductive state, since GLU and ASP were shown to be ineffective. Second, the competition of GLU and ASP with GABA for binding with the GLU-receptor indicates a significant contribution of γ-amino and distal carboxyl groups in the binding of the ligand to the receptor.

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