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ELECTROCARDIOGRAMS IN INSECTA AND GASTROPODA AND THEIR CHANGES UNDER EXPERIMENTAL CONDITIONS

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It was found that the electrocardiogram (ECG) in the Invertebrata is different in myogenic and in neurogenic hearts. Most of the authors concluded that the ECG of Mollusca is similar to the myogram of the smooth muscles (EVANS, 1912; ARVANITAKI and CARDOT, 1933; HENDRICKX, 1945; INOUE et al. 1950). In the EEG of Insecta slow as well as fast waves were separated of considerably different duration of cycle and amplitude (DUWEZ, 1936; CRESCITELLI and JAHN, 1938; PROSSER, 1950; MATSUI, 1955; IRISAWA et al. 1956). For a long time the shape of the ECG was taken as one of the basic criteria in the classification of the hearts and consequently, according to the origin of the rhythm the terms "myogenic" and "neurogenic" became generally used. Recently, however, some objections were raised against this distinction (MCCANN, 1970). This is the more justified, as the ECG of "myogenic" and "neurogenic" hearts was not studied with identical methods and so to emphasize the differences is not well-founded in all of the cases.

In our experiments the ECG of Gastropoda and Insecta hearts were compared as well as the effects of 5-hydroxytryptamine (5HT) and calcium ions on the ECG were studied.

Material and method

Experiments were carried out on the in situ hearts of *Helix pomatia* L. (Gastropoda) and of *Locusta migratoria migratorioides* R. F. (Insecta). The dissection and the perfusion of the hearts were made as had been described earlier (S.-Rózsa and GRAUL, 1964; S.-Rózsa and V.-Szőke, 1970). For recording ECG suction electrodes were used. In most of the cases both electrodes were placed to the surface of the ventricle (*Fig. 1.*) In some cases, however, one of the electrodes was put onto the auricle and another onto the ventricle. When the shape of the ECG was studied both of the electrodes were placed onto the auricle. In unipolar recording one of the electrodes was dipped into the surrounding fluid (*Fig. 1*). The tip of the suction electrodes varied between 1.0-2.5 mm. ECG registration was done with the standard photorecorder of the oscilloscope (Kardotester, Orion, Typ. 5101).

The substances were given to *Helix* by perfusion into the heart while to *Locusta* by change of the washing fluid to the surface. The experiments were performed at room temperature (20-24 °C).



Fig. 1. The location of the recording electrodes on the Helix heart A = auricle, V = ventricle

Results

The ECG of *Helix* heart can be registered in the form of bipolar or unipolar electrical signs characterized with some changes depending on the location of the recording electrodes.

a) In the case of auriculo-ventricular bipolar recording (Fig. 1) the ECG contained two fast waves of opposite direction. The direction of the potentials proved that the conduction of the impulses is carried from the auricle to the ventricle. The second fast wave always exceeded the first one. The fast components were accompanied by a slow negative wave (Fig. 2A). The most variable part is the slow wave differing from one heart to another in amplitude, duration and period as compared to the fast waves. The amplitude of the fast waves was altogether 18 ± 1.92 mV, the duration of a whole cycle varied between 120-250 msec.

b) When both leading electrodes were on the auricle the amplitude and the duration of the fast waves were identical with that of the previous registration, but their direction depended on the place of the electrodes. In most of the case the slow wave failed to appear or had positive value (Fig. 2B).

c) Unipolar registration. When one of the electrodes was dipped into the fluid a unipolar form of the ECG could be registered, whose amplitude was 10 mV with a duration of 250 msec (Fig. 2C).

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The fast waves appeared in each experiment with constant frequency simultaneously with the rhythm of the heart. Without external influences the ECG remained unaltered as long as 8-10 hours. However, considerable differences can be observed in the ECG of different hearts. In our experiments the frequency proved to be 24-48 cycle/min. The value of the amplitude was not influenced by the frequency. No relationship was observed between the duration and the frequency of the impulses for higher frequency was not accompanied by shorter duration.

The ECG of the Locusta hearts resembles in its shape to that of Helix and can be registered also as a bipolar or unipolar wave depending on the mode of the registration (Figs. 3 and 4). The values of the amplitude of the



Fig. 3. ECG of Locusta heart

- A. ECG from the second and fourth abdominal heart chambers
- B. ECG from the second abdominal heart chamber

C. Unipolar ECG

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Fig. 4. ECG from Locusta heart. Variability of the the slow waves

fast waves draw near $(18 \pm 2.44 \text{ mV})$ to that of *Helix*. The duration of the potentials was a little shorter in the *Locusta* heart: 100–180 msec. The frequency of the fast waves was 24–42 cycle/min. Also in this case the slow wave proved to be the more variable part of the electrical activity of the heart. In some cases the slow wave followed immediately after the fast ones but in other cases it was postponed in such a degree that seemed to precede the next fast wave (*Fig. 4A, C*). The variations of the slow wave could be observed on the same animal without any external influences. Occasionally in Insecta a second fast wave arose with lower amplitude (*Fig. 3A*). No oscillatory electrical changes were observed between the fast waves of the *Locusta* heart.

5-hydroxytryptamine (5HT) and alteration of the external calcium ion concentration proved to be very effective in influencing the heart rhythms as well as the ECG. The changes under these agents were estimated on the fast waves as it was satisfactorily stable on the control hearts.

Applying 5HT the rhythm as well as the frequency of the ECG increased on *Helix* heart. Simultaneously the amplitude of the fast waves decreased. The decrease of the amplitude was significant beginning from 10^{-8} M concentration of 5HT. 5HT increased the frequency by about 200 per cent at 10^{-6} M concentration (*Fig. 5*). Higher concentrations of 5HT failed to cause further increase in the frequency even very high concentrations $(10^{-3}-10^{-2} \text{ M})$ of 5HT decrease the frequency.

Having raised the concentrations of the external calcium ions both the amplitude and the frequency of the ECG showed some increase in *Helix* heart but this never exceeded the control value more than 25 per cent. In most of the cases the increase was only about 10 per cent.

In Ca^{++} -free solution an increase in the frequency and a decrease in the amplitude of the ECG was observed at the beginning (Fig. 5), then,







A. Control

- B. 6 minutes after Ca++-deprivation
- C. 7 minutes after Ca++-deprivation

after 5-10 minutes the arrest of heart activity took place. Preceding the arrest the first fast wave with negative direction was abolished, so the ECG became unipolar and deformed (*Fig. 6*). After washing the heart with physiological solution containing Ca⁺⁺ the original form of the ECG was restored. In Ca⁺⁺-free solution the ECG disappeared even in the case when 5HT was added in such a concentration which increased the frequency of the ECG in a normal physiological solution.

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In the Locusta heart the amplitude of the ECG was not influenced by 5HT, and also the frequency was increased only by 20 per cent. Increasing the external calcium concentrations the amplitude and the frequency of the Locusta ECG remained unaltered or increased only slightly. After withdrawal the Ca-ions from the medium both the slow and the fast waves of the ECG disappeared. After washing with normal physiological saline the original form of the ECG was restored in this species, too.

Discussion

According to our results the ECG of Helix and Locusta hearts are similar in form as well as in amplitude. In both cases unipolar or bipolar ECG can be registered depending on the mode of registration. The registered waves can be identified with the RS and T waves of the heart of Vertebrates. Our results did not proved the oscillatory nature of the ECG in Insecta hearts for between the fast waves no oscillatory electrical activity was observed. It seems probable that the oscillations reported earlier by many authors (DUWEZ, 1936; JAHN et al. 1937; CRESCITELLI and JAHN, 1938; MATSUI, 1955; IRISAWA et al. 1956) and regarded as a component of the ECG in Insecta hearts originate from the movement of the muscles fixing the heart. The above authors used not a suction but plating or silver electrodes for the leading off the ECG, thus the leading did not exclude even the possibility of the rise of rubpotential between the electrodes and the tissue moving rhythmically. The advantage of the method employed in our experiments is due to the fact that the electrical activity can be registered from a well determined area of the heart excluding the electrical activity of other tissues and preclude the possibility to registrate any rub-potential. In our case the Insecta ECG had a simple form and it cannot be compared with the complex electrical activity of the respiratory centres (PROSSER and BROWN, 1962).

The slow waves of the ECG both in *Helix* and *Locusta* hearts proved to be very variable, displaying permanent changes even under control conditions. For this reason the effect of the 5HT and Ca-ions was estimated only on the fast components. In case of *Helix* heart the amplitude of the fast waves was considerably affected by 5HT and in the Ca⁺⁺-free medium. The frequency of the ECG of *Locusta* heart was changed in a lower degree under the influence of 5HT but the omission of Ca-ions from the medium eliminated the ECG, like in *Helix* heart. The results showed that the fast waves can be influenced experimentally though the possibility of this was questioned earlier (SMIRNOVA and TURPAEV, 1948; DUWEZ, 1938). On *Helix* heart the lack of 5HT and Ca-ions effected the electrical activity of single myocardial cell and that of the total electrical activity in the same manner (KISS and S.-Rózsa, 1971), seemingly proving the existence of a connection between the excitatory effects of these factors. The presence of Ca⁺⁺-ions is necessary for the maintenance of the electrical activity and contraction of the heart in both species.

Our results emphasized that the shape and the character of the ECG cannot be taken as criteria for the classification of the hearts referring to the origin of their rhythm. Moreover, the existence of the oscillatory ECG characterizing the neurogenic hearts was not justified.

Summary

Electrocardiograms (ECG) were investigated using suction electrodes in Helix pomatia L. (Gastropoda) and Locusta migratoria migratorioides R.F. (Insecta). It was stated that by bipolar registration the amplitude of the ECG was nearly identical in the two species) Helix: 18 + 1.92; Locusta: 18 + 2.44 mV_{p-p}). The fast waves were also similar and it was registered in unipolar or bipolar form depending on the mode of the leading off. The fast waves were followed by a slow one, the latter being variable even under control conditions. The duration of the ECG was 120-250 msec on the Helix and 100-180 msec on the Locusta heart. The frequency of the ECG was 24-48cycle/min on the Helix and 24-42 cycle/min on the Locusta heart. Under control conditions the ECG of both species was rather stabile and it was maintained in an unaltered form for as long as 8-10 hours. Under the influence of 5HT the frequency of the ECG increased and the amplitude decreased. In the Ca⁺⁺-deprivation often an initial increase of the frequency and decrease of the amplitude was eliminated and the heart stopped. In Ca⁺⁺-excess a slight increase in the frequency and amplitude was observed.

The similar nature of the ECG registered from the two species showed that the shape of the ECG cannot be taken as a criterion for the classification of the heart in different animal phyla what would refer to the origin of the rhythm. The oscillatory electrical activity known as a component of Insect ECG is supposed to be of extracardial origin.

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GASTROPODA ÉS ROVAR SZÍVEK ELEKTROKARDIOGRAMJA ÉS ANNAK VÁLTOZÁSA KÍSÉRLETES KÖRÜLMÉNYEK KÖZÖTT

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Összefoglalás

Vizsgálták Helix pomatia L. (Gastropoda) és Locusta migratoria migratorioides R., F. (Insecta) szívének elektrokardiogramját (EKG) szívó elektródák alaklmazásával. Megállapították, hogy mindkét faj szívén közel azonos nagyságú (Helix: 18 ± 1,92, Locusta: 18 ± 2,44 mV_{p-p}) EKG regisztrálható bipoláris elvezetésben. Az EKG gyors komponensét is hasonlónak találták, s az elvezetés módjától függően uni- vagy bipoláris alakban regisztrálták. A gyors komponenseket egy lassú hullám követte, mely kontroll viszonyok mellett is változékonynak bizonyult. Az EKG időtartama Helix szíven 120 – 250. Locusta szíven 100–180 msec között váltakozott. Az EKG frekvenciája Helix szíven 24–84 c/min, Locusta szíven 24–42 c/min volt. Kontroll körülmények között mindkét faj szívének EKG-je állandó, mely változatlan formában fenntartható 8–10 órán át. 5HT hatására az EKG frekvenciája megnő, amplitúdója csökken. A Ca⁺⁺-megvonás esetén kezdeti frekvencia növekedés és amplitúdó csökkenés után az EKG eltűnik és a szívműködés megszűnik. Ca⁺⁺ hatására kisfokú frekvencia és amplitúdó növekedés jön létre.

A két faj szívén kapott azonos jellegű EKG arra mutat, hogy az EKG forma nem tekinthető elfogadható kritériumnak a különböző állatcsoportok szívritmusának felosztásában. A szerzők a korábban rovar szívek EKG-jében leírt oszcilláló elektromos aktivitást extrakardiális eredetűnek tekintik.