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Fish physiology

**EFFECTS OF ELECTRODE PLACEMENT RELATIVE TO THE CARDIAC
MUSCLE ON ECG PATTERN IN CARP (*CYPRINUS CARPIO* L.)**

**WPLYW TOPOGRAFII ELEKTROD WZGLĘDEM MIĘŚNIA
SERCOWEGO NA CHARAKTER ZAPISU EKG KARPIA
(*CYPRINUS CARPIO* L.)**

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Effects of electrode placement on the ECG quality were studied in 200–1500 g individual weight carp. Out of 12 combinations of lead placement relative to the cardiac muscle, 3 were found to produce the most legible, reproducible ECG's.

INTRODUCTION

Monitoring changes in fish electrocardiograms (ECG's) allows to collect information on, e.g., circadian rhythms (Węgrzynowicz et al. 1975), effects of anaesthetics and other chemicals (Serfaty et al. 1959; Muzykiewicz 1978; Mitsuda et al. 1980, 1988; Yamamitsu and Itazawa 1988a), effects of physical factors (Satchell 1961; Nomura et al. 1969; Wood et al. 1979; Yamamitsu and Itazawa 1988b, 1990), and behavioural traits (De Oliveira and Hoshino 1989).

Difficulties in obtaining an appropriate ECG in fish may stem from a small size of the heart, a low cellular membrane resting potential, a high skin resistance, and dispersion of electrical activity by water (Labat 1966; Ueno et al. 1986; Nanba et al. 1987; Satchell 1991).

The best ECG's in fish are usually obtained with a direct, invasive conduction of action potentials by inserting needle electrodes into the pericardium. The actual electrode placement depends on fish morphology. In the anguilliforms, the heart lies directly under

the abdominal covers, which makes it possible to use indirect conduction by inserting the leads just beneath the skin (Kłyszczko et al. 1977; Muzykiewicz 1978). In those fish equipped, like cyprinids are, with a well-developed pectoral girdle, the needle electrodes are inserted ventrally, between the cleithrum and the coracoid (Yamamitsu and Itazawa 1988a) or near the pectoral fins; the fish may be also “stitched through” with flexible leads (Labat 1966; Nanba et al. 1973; Yamamitsu and Itazawa 1988b).

However, ECG's obtained with all those methods are difficult to interpret because the leads are inserted deep into the fish body; the actual insertion is guided by the operator's eyesight only, with no guarantee that the lead will reach its destination near the heart. As a consequence, the ECG traces do not match, preventing comparability of the results. The ECG's may differ both in the shape and magnitude of waves.

The present work was aimed at finding a relationship between electrode placement and the resultant ECG pattern. In addition, a lead placement ensuring reproducible recordings of the carp heart electric activity without any need to open the abdominal cavity was sought.

MATERIAL AND METHODS

The study was carried out in winter. It involved a total of 36 carp of 200–1500 g individual weight, obtained from a cage culture situated in the heated water channel of the Dolna Odra power station near Szczecin. Before testing, the fish were acclimated in an a. 400 dm³ aquarium filled with aerated tap water at $19 \pm 1^\circ\text{C}$ and pH 7.2.

Each test was performed on one, randomly selected, individual. Before the test, the fish was anaesthetised for 10 min. with Propiscin [a pharmacological preparation developed exclusively for use, in immersion, in fish culture; the active substance is Etomidat, successfully used both in human and in veterinary medicine (Kazuń and Siwicki 1994; Trzebiatowski 1996)] administered at 2 ml/dm³ concentration.

The heart electric activity was monitored with an ASPEL AsCARD-31 electrocardiograph equipped with bipolar leads. The electrocardiograph had serially linked 35 and 50 Hz filters and an RS interface with a PC to process the data with a modified mathematical analysis software. All the ECG's were made at a constant paper strip movement rate (25 mm/s) and at a constant reference (1 mV = 10 mm).

Electric activity of the fish heart was recorded with an invasive method involving direct conduction of the potential difference. Two AgCl-covered silver, non-polarised, 3 cm long sharp-ended leads were used as electrodes. When making a recording, the procedure applied by Nanba et al. (1987) was followed, whereby the anaesthetised fish was placed, ventral side up, in a special stand to facilitate opening its abdominal cavity near the heart by making a 5–6 cm long longitudinal incision. A particular care was taken not to injure

the conus arteriosus and to retain the regular circulation. Subsequently, the two electrodes were placed near the heart, the third – grounding – one being inserted into the fish tail. The sequence of excitations was monitored on the screen. The electrocardiograph was set at recording Einthoven's first limbic conduction. Polarity of the leads was adjusted to obtain QRS waves directed upwards of the isoelectric line. The electrode connected to the positive electrocardiograph pole was placed near the atrium, the electrode connected to the negative pole being placed near the ventricle. The activity potential of the exposed surface of the heart was recorded in either manual or automatic mode.

RESULTS

The carp ECG's were obtained by placing the electrodes in 12 different positions relative to the heart muscle; they are shown in Fig. 1. The ECG components were found to differ, depending on the electrode placement, in the following way:

The atrial complex

The P wave was present in all the ECG's shown, its shape being strongly dependent on the presence of ventricular excitation, i.e., the V wave. The P wave amplitude was most often negative (electrode positions 1, 5, 10, and 11). It should be added that in each of the cases mentioned, the P wave was preceded by the V wave, reflecting the sinus venosus contraction. In three cases (positions 4, 6, and 12), a clearly positive P wave was recorded. The wave was poorly visible in two cases (positions 2 and 3), the very low amplitude being observed then.

The PQ segment: it was only in one of the ECG's shown (electrode position 11) that the segment was clearly visible and had a high amplitude; the segment's negative amplitude was observed with the electrode positioned as in 7 (Fig. 1).

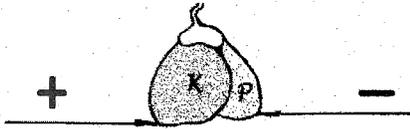
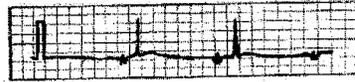
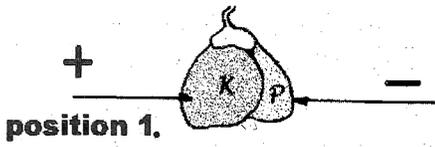
The ventricular complex

The Q wave was recorded in only two (positions 8 and 12) of the ECG's obtained, the wave missing in the remaining ones.

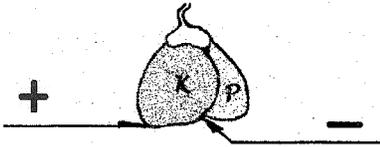
The R wave was present, with a positive amplitude, in all the 12 electrode positions.

The S wave was clearly visible on the ECG's obtained with positions 2, 7, 9, and 11 and was missing in positions 3, 5, 8, and 12.

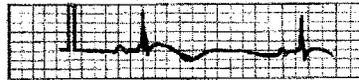
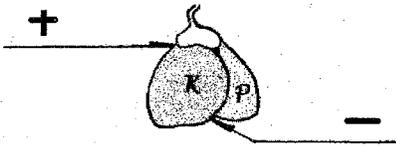
The T wave occurred, depending on the electrode placement, either above or below the isoelectric line. The positive T wave was recorded with electrode placed as in 1, 3, 5, and 6, the negative amplitude being visible in positions 4, 7, and 9. In those ECG's obtained with electrodes positioned in 2 and 8, the wave was missing altogether (Fig. 1).



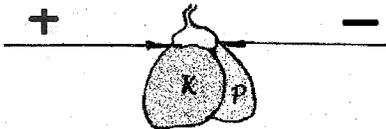
position 2.



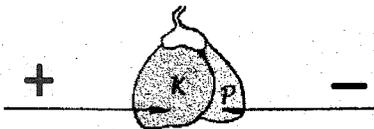
position 3.



position 4.



position 5.



position 6.

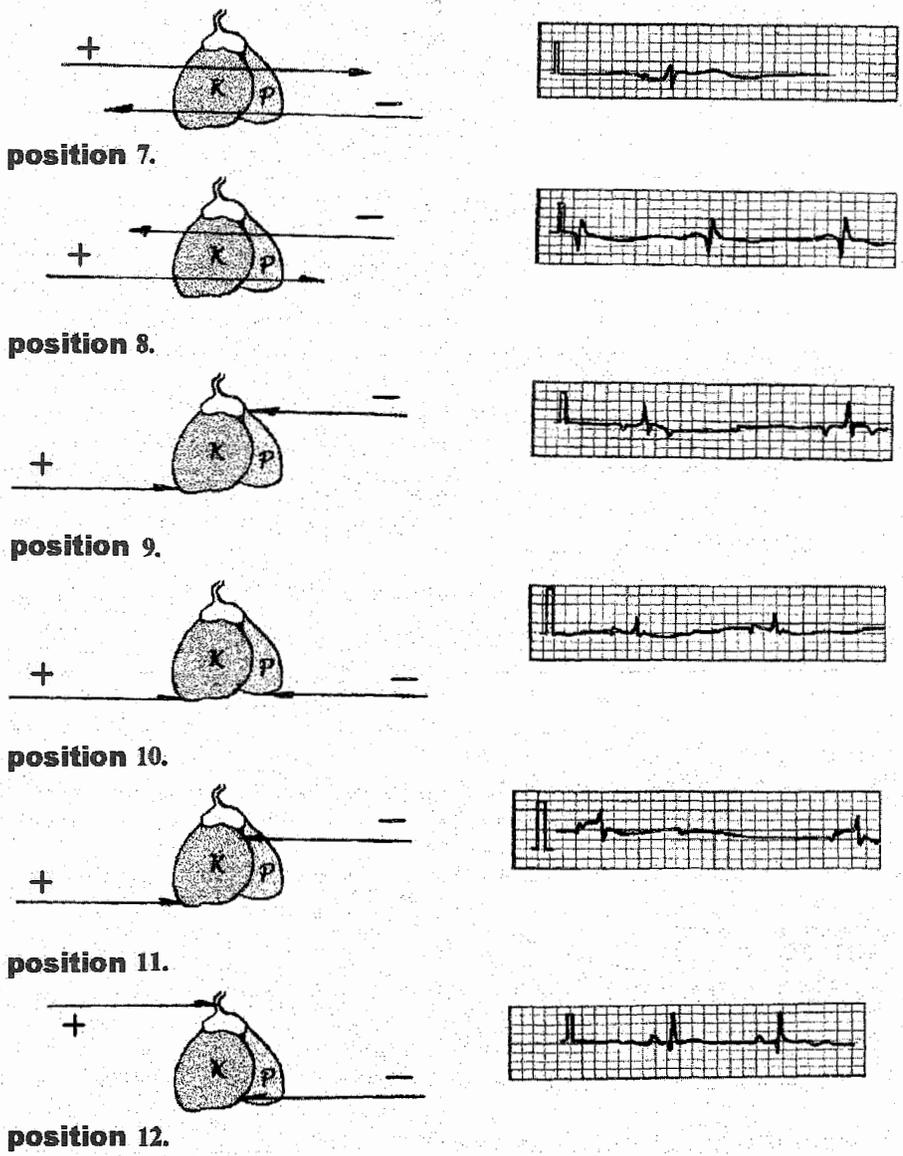


Fig. 1. The placement of electrode relative to the cardiac muscle and adequate ECG pattern; K, ventricle; P, atrium; +, positive electrode; -, negative electrode

DISCUSSION

The tests carried out on the open heart of the carp showed the carp ECG trace to have components similar to those visible in the human ECG. The typical P, Q, R, S, and T waves are present, which supports a number of earlier reports (Labat 1966; Nanba et al. 1973; Nanba and Murachi 1978; Ueno et al. 1986; Mitsuda et al. 1988). The carp ECG differs characteristically from the human one in, i.a., having a lower wave amplitude, due to the small size of the fish heart; displaying a highly irregular heart rhythm (within the physiologically sound limits); and showing the presence of the V wave, in the form of a sinusoid inflection preceding the P wave and interpreted as a reflection of the sinus venosus excitation (Satchell 1991).

Action potentials assume characteristic values in different places around the heart. The results obtained show all the 12 electrode positions used to be applicable in cardiac rhythm assessment. However, the most legible and reproducible ECG wave patterns were obtained with electrodes placed in positions 1, 6, and 12 (Fig. 1). With the electrodes placed in those positions, there is no need to open the fish body cavity for legible and reproducible ECG's to be obtained.

As shown by this study, the carp cardiac rhythm is highly variable and, like in some other fish species (Muzykiewicz 1978), may be arrested for a period of a few seconds to a few minutes.

CONCLUSIONS

1. A reproducible relationship between the electrode placement relative to the carp cardiac muscle and the ECG pattern was ascertained.
2. Positions of electrode insertion, most advantageous from the standpoint of obtaining reproducible ECG's without a necessity to open the body cavity, were located on the carp abdomen.

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WPLYW TOPOGRAFII ELEKTROD WZGLĘDEM MIĘŚNIA SERCOWEGO
NA CHARAKTER ZAPISU EKG KARPIA (*CYPRINUS CARPIO* L.)

STRESZCZENIE

Badano wpływ topografii elektrod względem mięśnia sercowego na charakter zapisu elektrokardiograficznego u karpia (*Cyprinus carpio* L.). Doświadczenia przeprowadzono na 36 sztukach karpia o masie jednostkowej od 200 do 1500 g. Przed każdym doświadczeniem rybę poddawano działaniu środka usypiającego przez okres 2 minut. Badanie elektrokardiograficzne przeprowadzono metodą ostrą, stosując odprowadzenia bezpośrednie dwubiegunowe, wzorowane na klasycznym odprowadzeniu II Einthovena.

W pojedynczym doświadczeniu zastosowano 12 kombinacji przyłożeń elektrod w poszczególnych miejscach wokół mięśnia sercowego.

Stwierdzono powtarzalną zależność pomiędzy charakterem zapisu EKG karpia i topografią elektrod względem mięśnia sercowego, a także ustalono najkorzystniejsze punkty wkłucia elektrod do powłok brzusznych karpia, pozwalające na uzyskanie powtarzalnych zapisów EKG bez konieczności otwierania jamy ciała.

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