Interaction of Inhibitory and Excitatory Junctional Potentials in the Control of a Myogenic Myocardium: the Ventricle of *Busycon canaliculatum*

Vagal and sympathetic stimulation have, respectively, hyperpolarizing and depolarizing effects on cardiac muscle cells of amphibia[1,2], reptiles[2], and mammals[3,4], but individual postjunctional potentials cannot be discerned in the hyperpolarizing action, and the depolarizing action appears as an increase in the slope of pacemaker potentials. In opisthobranch gastropods[5], individual facilitating and summating excitatory and inhibitory junctional potentials can be recorded from cardiac muscle. We wished to study in some detail the interaction of opposing junctional potentials in relation to contractile force of myogenic cardiac muscle. Some of our results have been reported in a brief abstract[6].

We studied cardiac muscle of the ventricle of a prosobranch gastropod, *Busycon* (Busycotypus) *canaliculatum* L. The methods were essentially the same as are being published elsewhere[7]. The cardiac nerve enters the ventricle embedded in the wall of the aorta. As it branches out across the ventricle the cardiac nerve remains embedded in a thin strand of muscle. Thus the 'nerve' is contractile and will retract if cut. In order to record externally from the intra-ventricular cardiac nerve, the muscle must be cut down. The intra-ventricular cardiac nerve was lifted on a pair of platinum wire electrodes for external recording. Stimuli were applied to the extra-ventricular cardiac nerve (proximal to the aorta) lifted on a pair of platinum wire electrodes. Conventional glass microelectrodes were used for intracellular recording from ventricular muscle and mechanical responses were recorded with an RCA 5734 mechano-electronic transducer.

Examples of hyperpolarizing responses and depolarizing responses are shown in Figure 1. Hyperpolarizing responses were more common. During repetitive nerve stimulation (visceral ganglion severed) potential difference (PD) gradually increased (Figure 1A). When stimulus intensity was increased (arrow) amplitude of compound nerve action potentials increased and the hyperpolarization was augmented. However, as high intensity stimulation was continued, depolarization began to prevail. Upon cessation of stimulation a local depolarizing response ensued. Such a depolarizing response can induce a mechanical contraction (Figure 1B). Presumably the local depolarizing response indicates that during the latter portion

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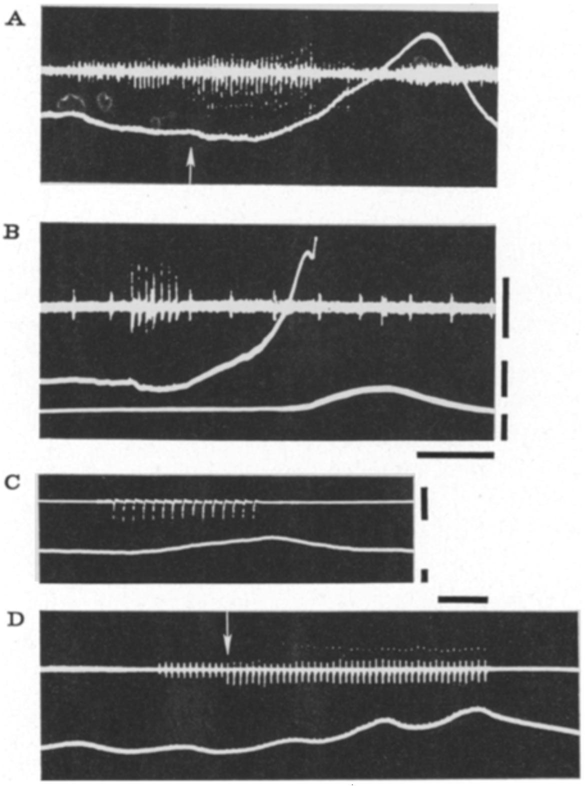

Fig. 1. A) Extracellular record from cardiac nerve (upper beam). Intracellular record from ventricular muscle (lower beam). B) Additional lowermost channel: tension. Calibration: 1 sec, 50 µV, 5 mV, 1 g. C) and D) Extracellular record from cardiac nerve (upper beam). Intracellular record from ventricular muscle (lower beam). Calibration: 1 sec, 50 µV, 3 mV.

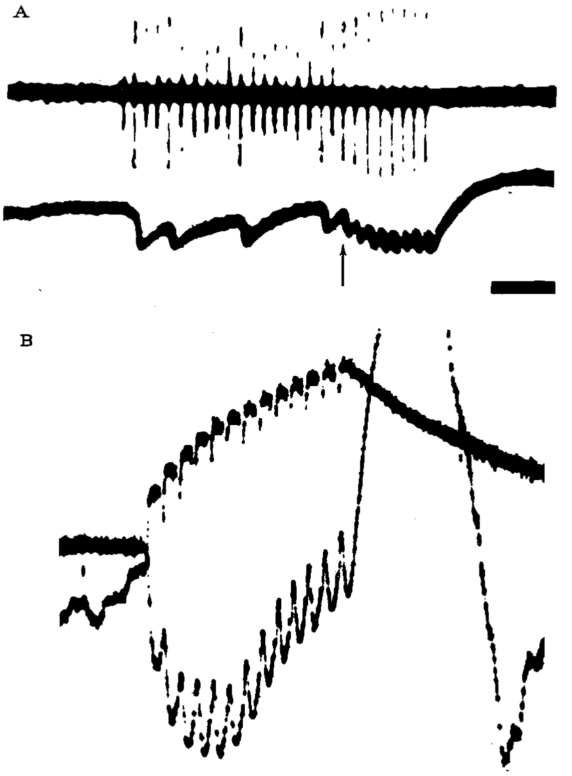

Fig. 2. A) Extracellular record from cardiac nerve (upper beam) and intracellular record from ventricular muscle (lower beam). Calibration: 2 sec, 100 µV, 1 mV. B) Upper beam indicates points of stimulation. Lower beam: intracellular record from ventricular muscle.

of the period of stimulation both inhibitory and excitatory nerve fibers were active and that rebound excitation followed the cessation of the inhibitory discharge. An example of a depolarizing response is shown in Figure 1C. During repetitive nerve stimulation (ganglion severed) depolarization gradually increased (Figure 1C). Spontaneous oscillation sometimes was apparent even before the beginning of nerve stimulation (Figure 1D). As depolarization grew, in response to repetitive nerve stimulation, the oscillation increased correspondingly in amplitude. At the arrow (Figure 1D) stimulus strength was increased. When the cardiac nerve was stimulated just at inhibitory threshold intensity, compound nerve action potentials appeared, of intermittently fluctuating amplitude, as in the first part of the record in Figure 2A. The largest of these nerve action potentials induced unitary inhibitory junctional potentials. When suprathreshold stimulation was given (arrow) repetitive large compound nerve action potentials evoked repetitive summating inhibitory junctional potentials. These may hold the membranes hyperpolarized.

In Figure 2B, we see that rebound excitation from inhibitory junctional potentials and usual excitation from excitatory junctional potentials can occur at the same time. Nerve stimulation first induced hyperpolarization, but then depolarization prevailed. An action potential followed cessation of stimulation. It may be thought that this kind of overlapping action causes especially rapid and strong excitation[8].

Résumé. Les auteurs ont étudié, par microélectrode interne, l'interaction entre les potentiels postsynaptique inhibitoire et excitatif dans un myocarde myogénique. Ils ont observé la sommation de ces potentiels et leur action facilitant l'inhibition par hyperpolarization et l'excitation par depolarisation.

K. Kuwasawa[9] and R. B. Hill

Zoological Institute of the Tokyo Kyoiku University,
Tokyo (Japan), and Department of Zoology, University
of Rhode Island, Kingston
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[9] Address: Zoological Institute, Faculty of Science, Tokyo Kyoiku University, Tokyo (Japan).

Decreased Blood P_{CO_2} in the Ovine Foetus During Hyperthermia: Implications for Increased Placental Blood Flow

During experiments designed to examine factors determining the onset of parturition and the time at which the foetus first becomes thermosensitive, we have observed that thermally-induced hyperventilation in the ewe caused a far greater depression of P_{CO_2} in foetal, as opposed to maternal blood; a likely explanation for this is that placental blood flow increased.

It has recently been shown that in the conscious sheep, the difference between maternal and foetal blood P_{O_2}, P_{CO_2} and pH remained virtually constant during small fluctuations in these parameters[1]. However, large changes in blood gas status of the ewe induced by artificial hyperventilation or by administration of gas mixtures, resulted in wide variations in the materno-foetal blood P_{O_2} difference, whereas the P_{CO_2} difference remained essentially constant. These latter studies were performed either with the ewe under general anaesthesia and the foetus in utero[2], or with the ewe under spinal anaesthesia and the foetus partly exposed[3].

Methods. One Merino × Merino (sheep No. 1) and 2 Border Leicester × Merino (sheep Nos. 2 and 3) foetuses were surgically perpared and maintained with a vinyl catheter (1 mm i.d., 2 mm o.d.) in the left common carotid artery as described by Bassett, Thorburn and Wallace[4]. At the same operation a copper-constantan thermocouple (38 swg.) clad in vinyl was sutured into the sheath of the right common carotid artery of the foetus and ewe to permit the monitoring of foetal and maternal arterial blood temperatures ($T_{ar'}$ ±0.05°C). Blood samples (2 ml) were drawn into chilled, heparinized syringes. The P_{O_2}, P_{CO_2} and pH were measured at 38°C using a Radiometer BMS 3 electrode system, and corrected to the animals' temperature with appropriate factors[5,6], shown to be applicable to the sheep[7]. Oxyhaemoglobin saturation (S_{O_2}) was measured using an Instrumentation Laboratory CO-Oximeter (Model 182) calibrated for use with sheep blood[8].

Animals were housed together in a climatic room with an ambient dry bulb temperature of approximately 16°C. Observations were made on sheep Nos. 2 and 3 during the last 16 days of the 150 days gestation period. A moderate heat stress was imposed between day −14 and −10 of pregnancy (day 0 is day of birth) by raising the ambient temperatures to 40°C dry bulb and 26°C wet bulb. Temperatures of the ewe and foetus were recorded continuously for 1 h periods in the early morning, midday and late afternoon of every day; a blood sample was drawn from the foetus during each morning period. Sheep No. 1 was studied from day −26 but observations relevant to the present report were made only on day −25. This preparation had additional catheters, 1 in an umbilical vein of the foetus and 1 in a maternal common carotid artery. On day −25 temperatures were monitored continuously for $1^{1}/_{2}$ h before, 3 h during and 2 h after exposure to moderate heat stress; 2 blood samples were taken before and after, and 3 during heat exposure.

Results and discussion. Individual results for each animal are summarized in the Table, wherein mean values are given for the various parameters during the control periods (pre- and post heating) and during heat exposure. Foetal T_{ar} was normally 0.4–0.5°C higher than maternal

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