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CHOLINESTERASE ACTIVITY OF LEFT AND RIGHT ATRIA OF THE RABBIT'S HEART

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Rhythmic activity in the atria of the rabbit's heart has been shown to be closely linked with the synthesis of acetylcholine within the tissue (Bülbring & Burn, 1949). Observations by Burn & Kottogoda (1953) on the action of eserine on rabbit atria supported this hypothesis and suggested that not only the synthesis of acetylcholine but also its destruction by cholinesterase were important for the maintenance of normal activity. Bülbring, Kottogoda & Shelley (1954) estimated the cholinesterase activity in homogenates of rabbit atria and found a correlation between this and the sensitivity to eserine of atria beating in corresponding conditions. The action of other anticholinesterases, including neostigmine and diisopropylfluorophosphonate, in low concentrations also supported the view that cholinesterase activity in atria was important for the destruction of acetylcholine synthesized in the tissue during its rhythmic contraction (Briscoe & Burn, 1954).

All the work referred to above was concerned with a single preparation of both left and right atria. Although Brouha & Bacq (1936) state that all cardiac tissue, including the left atrium of the rabbit, beats spontaneously if perfused with an adequately oxygenated physiological solution, many workers have found that the isolated mammalian left atrium does not contract rhythmically except in rare instances, although it may beat in the presence of adrenaline or histamine (Rigler & Tiemann, 1929; Kruta, 1934; Rothberger & Sachs, 1939). It will also beat rhythmically in response to mechanical or electrical stimulation, the response outlasting the stimulation (Hermann, Cornut & Guiran, 1937; Segers, 1947). Thus although the left atrium possesses the property of rhythmic movement when stimulated, the initiation and regulation of the beat may be a property of nodal tissue in the right atrium. If this is so, the cholinesterase activity of the right atrium might differ from that of the left.

Antopol, Glaubach & Glick (1939) found that the left atria of rabbits had a slightly higher cholinesterase activity against acetylcholine than the right

atria. These workers made no attempt to distinguish between true and pseudo-cholinesterase activity. The activities of both true and pseudo-cholinesterase in the left and right atria of rabbits have now been determined by the use of the specific substrates acetyl- β -methylcholine (MCh), benzoylcholine (BCh) and butyrylcholine (BuCh). Acetyl- β -methylcholine is hydrolysed by true but not by pseudo-cholinesterase, whilst benzoylcholine is attacked by pseudo- but not by true cholinesterase (Mendel, Mundell & Rudney, 1943). Butyrylcholine is rapidly hydrolysed by pseudo-cholinesterase, but is hardly attacked by true cholinesterase (Nachmansohn & Rothenberg, 1945).

METHODS

The atria from freshly excised rabbit hearts were dissected free from fat, connective tissue and ventricular muscle. The left and right atria were separated by a cut to the left of the interauricular septum.

In each experiment two left atria were dried on filter paper, and weighed together, and two right atria were similarly dried and weighed. Each pair of atria was then chopped on filter paper on the stage of a mechanical chopper (McIlwain & Buddle, 1953). The mince obtained was homogenized in Krebs-Henseleit bicarbonate solution. Usually 4 ml. of this solution per g of atrium was used, but if the atria were very small, 5 ml. was used per g atrium. Care was taken that in each experiment the concentrations of the left and right atria homogenates were identical.

Cholinesterase activity was determined manometrically by the standard Warburg procedure at 37.5° C. Owing to the small amount of material available, micro-flasks were used. The following substrate solutions were made up in distilled water:

Acetylcholine (acetylcholine chloride, Roche Products Ltd.), 0.2M.

Acetyl- β -methylcholine (Amecho, Savory and Moore Ltd.), 0.3M.

Benzoylcholine (Benzoylcholine chloride, B.D.H. Ltd.), 0.06M.

Butyrylcholine (Butyrylcholine chloride, B.D.H. Ltd.), 0.2M.

0.1 ml. of substrate solution was placed in the side bulb.

0.5 ml. homogenate was placed in the main compartment, and the volume was made up to 0.8 ml. with Krebs-Henseleit solution.

Five-minute readings were taken over the first 33 min after adding the substrate, and the activities were calculated from the initial slope of the graph of carbon dioxide evolved against time.

RESULTS

The results of eleven experiments are shown in Table 1. Cholinesterase activity is expressed as μ l. CO₂ evolved per g atrium per hr. When acetylcholine was used as substrate, the total cholinesterase activity was very similar in left and right atria, the mean activity of left atria being only slightly higher than the mean activity of right atria.

The true cholinesterase activity, as measured by the hydrolysis of acetyl- β -methylcholine, was higher in the right than in the left atrium in nine out of ten experiments. This difference in activities was significant ($P < 0.001$). The pseudo-cholinesterase activity was, however, higher in the left atrium than in the right in each of ten experiments. The rate of hydrolysis of benzoylcholine by the left atrium was significantly higher than by the right ($P < 0.02$) and the hydrolysis of butyrylcholine was also significantly higher ($P < 0.001$).

TABLE 1. Cholinesterase activity in $\mu\text{l. CO}_2/\text{g atrium/hr}$

Expts.	Right atria				Left atria			
	ACh	MCh	BCh	BuCh	ACh	MCh	BCh	BuCh
1	1050	—	—	—	731	—	—	—
	938	—	—	—	638	—	—	—
2	1155	225	120	—	840	85	165	—
3	881	281	56	—	1181	300	250	—
4	950	220	128	—	1280	146	275	—
5	1950	300	375	—	2190	210	555	—
6	1260	375	225	—	1200	225	285	—
7	1710	350	—	2460	1830	270	—	3525
8	1470	360	—	1980	1560	225	—	2550
9	1530	390	—	2070	1455	270	—	2610
10	1660	542	—	1860	1525	253	—	2400
11	2130	360	—	2700	2790	315	—	3750
Mean	1390	340	181	2214	1435	230	306	2967

DISCUSSION

The fact that the right atrium of the rabbit has a higher true cholinesterase activity than the left atrium may be associated with the greater concentration of nodal tissue in the right atrium. The sino-auricular node is surrounded by a rich plexus of nerve endings (Flack, 1910), and nervous tissue is known to contain mainly true cholinesterase (Nachmansohn & Rothenberg, 1945). However, nerve endings and ganglia are by no means confined to the right atrium (Woollard, 1926), and the higher true cholinesterase activity of this atrium may not be connected with nervous elements.

SUMMARY

1. The cholinesterase activity of the left and right atria of the rabbit's heart has been determined manometrically, using specific substrates for true and pseudo-cholinesterase.
2. True cholinesterase activity is higher in the right atrium than in the left. Pseudo-cholinesterase activity is higher in the left atrium than in the right.

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