Effect of Angiotensin II on Neonatal Lamb Carotid Arteries[1]

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Summary. Isometric tension was measured in arterial strips from neonatal lambs and adult sheep, after stimulation by angiotensin II. During the early maturation period immediately following birth (3 weeks) there was a progressive increase in sensitivity to the agent.

The mammalian cardiovascular system is characterized by rapid anatomical and functional development during the early neonatal period. Functional aspects have been investigated mostly from the point of view of determining the level of reflex control in the newborn (MOTT[2]; SHINEBOURNE et al.[3]) and in general, reports indicate that the adrenergic and cholinergic receptor systems, both neural and humoral, are present at birth, but that they operate at a lower level than in the adult. The response characteristics to other vasoactive agents have not been studied as extensively, so the present in vitro study attempts to describe the maturation of vascular reactivity to angiotensin in neonatal lambs during the immediate postnatal period.

Methods. Carotid arteries from neonatal lambs and adult ewes (Group I: up to 24 h old; II: 2–8 days old; III: 14–22 days old; IV: adults) were cut into helical strips, 2 cm long by 2 mm wide, and suspended under tension in a 10 ml organ bath of Krebs bicarbonate solution bubbled with 95% O_2/5% CO_2. A force transducer above the bath measured developed (isometric) tension in the strip. After 90 min of equilibration angiotensin (conc. range of 2.5×10^{-7} to 8×10^{-3} mg/ml) was added to the bath in 0.05 ml volumes, in cumulative steps. Log dose-response curves were plotted from the data and threshold (conc. producing a 2% maximal response) and ED_{50} (concentration corresponding to the half-maximal response) were determined. 21 animals were studied with 2–4 strips/animal used in each study.

Results. Vascular strips from animals in all groups (I, II, III, IV) responded to the addition of angiotensin by contraction. The data from the log dose-response curves are compiled in Table I and in the Figure (Mean and SEM for threshold, ED_{50}, and maximal tension). With increasing age there is an increase in sensitivity of arterial smooth muscle to angiotensin, as indicated by the steady decrease in threshold and ED_{50} (except for a slight increase in ED_{50} from group II to group III, which is not significant) from the day of birth up to the adult stage. During the 3 week neonatal period under study the threshold decreased by 76.5% and the ED_{50} by 70.8%. In that same period the maximal tension which the strips were able to develop more than doubled. Following that 3 week period there were further changes which occurred before adult values were attained, the threshold decreased by 70%, the ED_{50} by 48%, and there was a further 1.8-fold increase in maximal tension.

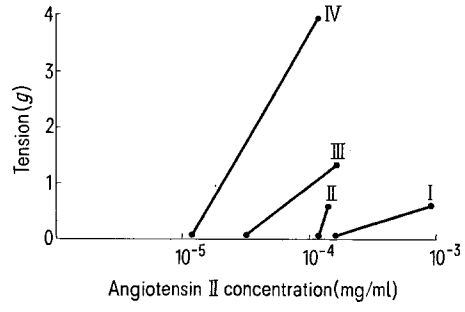
For purposes of comparison, data from another study on adrenergic responses in newborn lambs is included here. In Table II response data for norepinephrine are compiled, and it can be seen that there is also a progressive increase in developed tension (1.3-fold) and a steady decrease in threshold (81.6%) and ED_{50} (72.4%) values during the first 3 weeks of life. However, there was very little change in reactivity after the 3 week period (although maximal tension continued to rise), contrary to what is seen with angiotensin which continues to change.

Table I. The effect of angiotensin II on arterial strips from neonatal lambs and adult sheep (Mean + SEM)

Group	Threshold (mg/ml)	ED_{50} (mg/ml)	Maximal tension (g)
I	$2.36\pm0.15\times10^{-4}$	$9.16\pm0.44\times10^{-4}$	1.28 ± 0.33
II	$1.62\pm0.10\times10^{-4}$	$2.30\pm0.27\times10^{-4}$	1.39 ± 0.07
III	$5.25\pm0.20\times10^{-5}$	$2.97\pm0.64\times10^{-4}$	2.73 ± 0.61
IV	$1.57\pm0.55\times10^{-5}$	$1.53\pm0.39\times10^{-4}$	7.88 ± 1.18

Table II. The effect of norepinephrine on arterial strips from neonatal lambs and adult sheep (Mean ± SEM)

Group	Threshold (mg/ml)	ED_{50} (mg/ml)	Maximal tension (g)
I	$6.00\pm1.35\times10^{-5}$	$6.47\pm0.96\times10^{-4}$	2.02 ± 0.34
II	$1.59\pm0.50\times10^{-5}$	$2.55\pm0.44\times10^{-4}$	2.57 ± 0.35
III	$1.10\pm0.24\times10^{-5}$	$1.78\pm0.13\times10^{-4}$	4.63 ± 0.73
IV	$1.10\pm0.05\times10^{-5}$	$1.76\pm0.12\times10^{-4}$	10.40 ± 1.48



Effect of angiotensin II on carotid artery strips of neonatal lambs and adult sheep. The line for each group connects the threshold concentration (point near the x-axis, tension is only 2% of maximal) and ED_{50} (concentration corresponding to the half-maximal tension). There is a steady decrease in both values and a consequent increase in the slope of the lines with age (except for the large increase in group II). I, first 24 h; II, 2–8 days of age; III, 14–22 days of age; IV, adult ewes.

[1] This study was supported by the Golden Empire Chapter of the American Heart Association and the United States Public Health Service, grant No. PHS HL 14780-03.

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Discussion. The data indicate that there are significant changes in arterial responsiveness to angiotensin II during the early neonatal period in lambs, just as has been found with α-adrenergic agents (KNIGHT and MCGREGOR[4]; DE CHAMPLAIN et al.[5]; GRAY[6]). The smooth muscle cells become more sensitive to this polypeptide molecule during early maturation, although at the end of the end of the 3 week period under study, the vessels are still less sensitive than comparable vessels from the adult animal and they are capable of developing only about $^1/_3$ the maximal tension. In contrast, norepinephrine sensitivity changes drastically during the 3 week period but does not change very much after that, even though there is still a large change in tension occurring after that period. In the Figure there is a progressive change in slope (except for group II which showed a large change in slope) of the curves depicting the relationship between threshold, ED_{50}, and tension. This is an indication that the threshold concentration (which stimulates the most responsive cells in the population) changes at a slightly different rate than the ED_{50} (which is related to the reactivity of the majority of cells) during the process of maturation, even though both are decreasing.

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The Effect of Intraventricular Thyroxine Administration on Body Temperature in Dogs at Rest and During Physical Exercise

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Summary. Infusion of 1 μg thyroxine into the left cerebral ventricle of the dog did not change body temperature at rest, but it caused significantly higher increases in T_{re} during physical exercise.

It has been found in this laboratory[1] that in dogs with excess of thyroid hormones temperature, responses to exercise are much higher than in control animals. It was also demonstrated that the thyroid-hormone-induced exercise hyperthermia cannot be attributed exclusively to the metabolic heat production[2]. Another possible explanation of this phenomenon is that thyroid hormones exert their action on central nervous structures involved in temperature regulation. An effect of thyroxine on the thermoregulatory centres has recently been described in the cat[3].

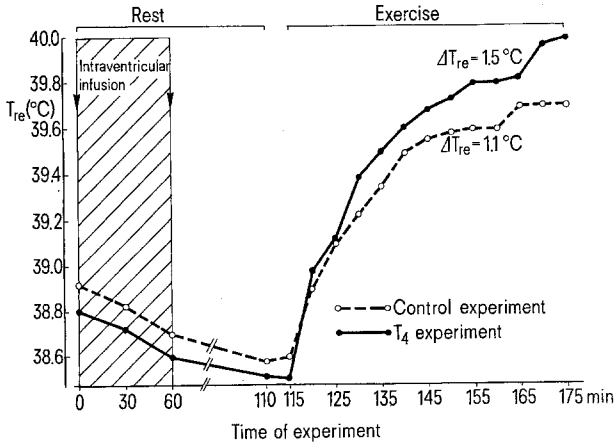
The purpose of the present work was to follow up the effect of intraventricular infusion of thyroxine on deep body temperature in dogs at rest and during physical exercise.

Material and methods. 6 mongrel dogs (18–22 kg b.wt.) were used. They were deprived of food for 18–20 h before the experiments, but had free access to water. At least 2 weeks before the experiments started each dog had a stainless steel guide cannula implanted into the left lateral ventricle. The operation was performed under Na-heksobarbital anaesthesia (45 mg/kg i.v.).

Control experiments. Dogs trained to stand quietly on a stand were infused intraventricularly with the artificial cerebrospinal fluid (ACSF) at a rate of 40 μl/min for 60 min. After the end of the infusion, the dogs remained on the stand for a further 50 min. Then, they were transferred to an electrical treadmill, where they began a 1 h physical exercise (slope of treadmill of 12°, speed 1.2 m/sec). A thermocouple (Ellab, Copenhagen) was inserted 12 to 15 cm deep into the rectum. Rectal temperature was read every 2 min throughout the experiment, and at 5 min intervals after termination of exercise until T_{re} returned to the pre-exercise value.

Experiments with thyroxine were performed on the same dogs according to the scheme described above, but thyroxine (Light and Co, England) in a total amount of 1 μg/dog was added to the ACSF infused.

In both series of experiments, venous blood samples were taken at the beginning of infusion, immediately before exercise, and after its termination, for hematocrit and plasma free fatty acid (FFA) level determinations[4,5]. The data were analyzed with the Student's *t*-test for paired samples.



Effect of intraventricular infusion of 1 μg thyroxine on rectal temperature (T_{re}) of a dog at rest and during 1 h physical exercise.

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