

SPECIFIC AND SEASONAL VARIATIONS IN DEVELOPMENT OF DIVING BRADYCARDIA IN ANURAN AMPHIBIA

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Abstract—1. Submersion causes bradycardia, during November–December, in *Rana pipiens*, *R. temporaria*, *Bufo bufo* and *Xenopus laevis*. Heart rate stabilizes after 30 min submersion. The underwater stabilized rate is similar in all species except *X. laevis*.

2. Adjustment from surface to underwater stabilized heart rate occurs most rapidly in *R. pipiens*.

3. Rate of adjustment is slower and underwater equilibrium heart rate higher in summer *R. temporaria* and *B. bufo* as compared with winter and breeding animals.

4. During hibernation of *R. temporaria* heart rate is low and similar in animals of a wide size range.

5. Both seasonal and specific differences are discussed in terms of variations in nervous and metabolic activity.

INTRODUCTION

IN DIVING vertebrates bradycardia is the most obvious manifestation of a series of cardiovascular adjustments which occur during submergence. Bradycardia may also occur in strictly terrestrial vertebrates during periods of apnoea, but reduction in heart rate is usually considerably smaller than occurs in well-adapted divers (Bond *et al.*, 1961; Scholander, 1962). Anuran amphibia can respire cutaneously when pulmonary ventilation ceases but, in spite of this, oxygen lack is the major cause of diving bradycardia in *Rana temporaria* (Jones & Shelton, 1964). Decrease in heart rate is a reflection of the animal's inability to extract sufficient oxygen from the medium to maintain normal metabolism, because when *R. pipiens* is submerged in water of $pO_2 = 760$ mm Hg oxygen consumption remains at the surface rate and bradycardia does not occur (Jones, 1967). During the early part of submergence, oxygen is probably withdrawn from the lungs to supplement that obtained cutaneously; nevertheless arterial oxygen tension falls rapidly in *R. catesbeiana* (Lenfant & Johansen, 1967). Gas exchange during prolonged submergence is solely cutaneous so that factors affecting cutaneous oxygen exchange may be reflected in the development of diving bradycardia.

Rate of gas exchange across an unventilated but vascularized surface will depend on the metabolic demands of the animal which in turn may be limited by

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the nature of the surface for gas exchange (area, thickness, etc.) and the characteristics and time of exposure of the blood to the external medium. Consequently, variations in any of these factors may offset or enhance diving bradycardia of amphibia due to changes in the capacity for cutaneous gas exchange.

Seasonal variations in amphibian respiratory metabolism are well documented (Krogh, 1904; Dolk & Postma, 1927). Not only does total gas exchange vary but also the relative importance of lungs and skin. However, in the presence of pulmonary respiration cutaneous oxygen uptake remains more or less constant throughout the year (Krogh, 1904) but in curarized *R. pipiens* (completely immobilized) oxygen consumption is maximal in spring and minimal in winter (Fromm & Johnson, 1955). The density of cutaneous vasculatization of amphibia appears to be constant throughout the year (Czopek, 1958; Poczopko, 1958), but the number of capillaries in the web of *R. esculenta* which open during respiratory stress varies seasonally (Poczopko, 1958). Such cutaneous vasomotor reflexes are strongest during the breeding season and weakest during autumn. Furthermore, the number of erythrocytes, blood volume and haemoglobin content of blood also vary, being lowest during the winter months (Heesen, 1924; Holzapfel, 1937). As well as seasonal variations specific differences in respiratory metabolism, skin vasculatization and blood characteristics occur in anuran amphibia. Oxygen uptake of mature *Xenopus laevis* (aquatic) is considerably lower than that quoted for more terrestrial anurans (Charles, 1931; Jones, 1967). The skin of *Bufo bufo* is thicker and less well vascularized than that of aquatic anurans (Czopek, 1955). McCutcheon & Hall (1937) have also shown that the oxygen dissociation curve of the blood of aquatic amphibians is not sigmoid but tends to a rectangular hyperbola. The purpose of the present investigation was to determine whether variations of this type, both seasonal and specific, were reflected in the diving performance of several species of anuran amphibia, with respect to reduction in heart rate during submergence.

METHODS

Experiments have been performed on fifty-four *Rana temporaria*, twenty-one *R. pipiens*, nine *Xenopus laevis* and fifty-five *Bufo bufo*. All experiments were carried out at $17 \pm 0.5^\circ\text{C}$ except hibernation experiments which were done at $4-5^\circ\text{C}$. Seasonal animals were obtained direct from suppliers, their previous history being unknown. They were kept in the laboratory, under a natural photoperiod, at $17-18^\circ\text{C}$ for a period not exceeding 3 weeks, for any one individual, before the experiments and were offered blow-fly larvae. Feeding has apparently no effect on oxygen consumption, carbon dioxide production or respiratory quotient (Fromm & Johnson, 1955).

Heart rate was recorded by thin copper wire sewn into the chest, the indifferent electrode being at earth. The signal was amplified by conventional means and displayed either on an A.E.I. pen recorder or Sanborn 966-6 direct pen writer. Breathing movements of winter animals only were recorded by a photocell as described previously (Jones & Shelton, 1964; Jones, 1967).

The animals were attached to a weighted cork platform and allowed 1-2 hr acclimatization, at the surface, before the start of the experiment. An initial recording of heart rate was taken and the animal was submerged in a box of 12-l. capacity, the water being unstirred.

After 1 hr the animal was allowed to surface. To aid acclimatization and avoid disturbance of the animal the experiments were performed in a darkened room. Experiments on hibernation were carried out from December to January by keeping six *R. temporaria* of various sizes at $+5^{\circ}\text{C}$; they were placed in large jars, 75 cm high, containing 6–7 l. of water. Moss or leaf mould was placed in the bottom of the jars so that the animals could burrow. Frogs were not denied access to the water surface. In all experiments recovery was complete, most of the animals surviving for a period of 3–6 months after this series of experiments. Results were analysed statistically by means of Fisher's *t*-test and 1 per cent was considered the fiducial limit of significance.

RESULTS

1. *Specific variations between R. pipiens, X. laevis, R. temporaria and B. bufo*

(a) *November–December*. Animals were examined during November and December of 2 years. During this time a period of quiescence may occur in the wild, i.e. hibernation in *R. pipiens*, *R. temporaria* and *B. bufo*, aestivation in *X. laevis* (Alexander & Bellerby, 1938). The latter were shipped from South Africa and had probably been under controlled conditions for a period of 5–6 weeks longer than the other species studied at this time.

Submersion caused bradycardia in all species (Fig. 1). The period of submergence necessary to cause a fall in heart rate was frequently above that at the surface. Many animals made buccal breathing movements during the early stages of submersion; air was withdrawn from the lungs and forced back in by a rapid lowering and raising of the floor of the buccal cavity. Air was occasionally lost during these manoeuvres. For convenience the curves shown in Fig. 1 were divided at the 30-min mark. The curves were compared statistically during the period when heart rate was falling (0–30 min submergence) and after heart rate stabilized (35–60 min) (Fig. 1). A comparison between *X. laevis* and the other species, during the falling phase, could not justifiably be made since the surface heart rates were very different. Decrease in heart rate between the second and thirtieth minutes of submersion in *R. pipiens*, *R. temporaria* and *B. bufo* was significantly different in each species. An approximate analysis of the actual rate of adjustment to underwater equilibrium using the mean values for each species (Fig. 2) showed that adjustment was most rapid in the case of *R. pipiens*, being about 1.5–2 times as fast as the other species (Fig. 2). When drawn for individual frogs, curves of the type shown in Fig. 2 were complicated by the existence of a variable period before the drop in heart rate began; this made it difficult to make an accurate assessment. The difficulty was most acute in the case of *R. temporaria*. Consequently an attempt was made to fit exponential decay curves of the type

$$y = a + b e^{-kt}$$

for each individual. The exponent K gave a measure of the speed of adjustment, since when time $t = 0$, heart rate $y = a + b$ and when $t = \infty$ then $y = a$. The results agreed with the slopes of logarithmic transformation shown in Fig. 2 in the general picture but not precisely in detail. Nevertheless, the existence of a genuine

specific difference between the response of *R. pipiens* ($K = 0.18$) and the others ($K < 0.1$) was confirmed.

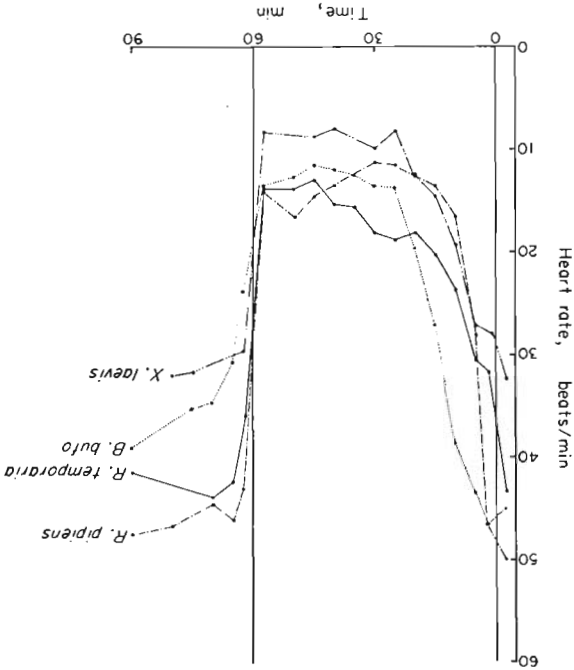


FIG. 1. Bradycardia caused by submergence and recovery on surfacing during November–December. *R. pipiens* (10 experiments, av. wt. 37.25 ± 1.5 g). *R. temporaria* (8 experiments, av. wt. 43.6 ± 3.0 g). *B. bufo* (8 experiments, av. wt. 45.8 ± 1.5 g). *X. laevis* (9 experiments, av. wt. 39.5 ± 4 g). Submersion and emersion at 0 and 60 min respectively.

Rapid fall of heart rate of *R. pipiens* was usually associated with heart beat becoming arrhythmic. After 10 min submersion 80 per cent of these frogs had developed arrhythmia. In the other species peak numbers of animals showing arrhythmia occurred somewhat later (15–25 min). After 20–35 min submersion rhythmic contractions were generally restored and rate of heart beat remained low. Early occurrence of arrhythmia was generally associated with more rapid adjustment of heart rate to underwater equilibrium in all animals. Figure 3 illustrates this relationship for every animal reported in this paper.

In some members of all species arrhythmia occurred during the later stages of submersion and this was most common in *R. pipiens*. A few animals performed buccal movements after establishment of the underwater equilibrium heart rate; these were of small amplitude and appeared to have no effect on heart rate. The stabilized heart rate was lower in *X. laevis* than the others by some 4–6 beats/min, which was significantly different from the other species. The surface rate of *X. laevis* was also below that of the other species (Fig. 1). There was no significant

Fig. 3. Relationship between rate of adjustment to underwater stabilized heart rate and the time at which pronounced arrhythmic heart beat first occurred. Submersion at 0 min.

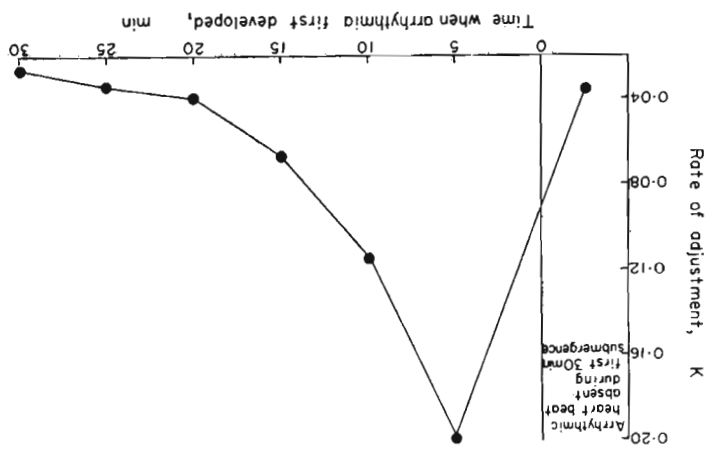
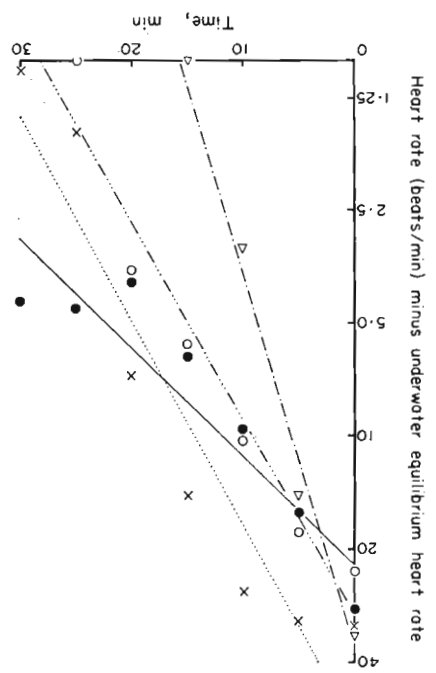


Fig. 2. Rates of adjustment of heart rate to the underwater equilibrium during the first 30 min of submergence. November-December, same data as Fig. 1. *R. pipiens*: \times , *B. bufo*; \circ — \circ , *X. laevis*; \bullet — \bullet ; *R. temporaria*; \triangle — \triangle , *R. pipiens*. Submersion at 0 min. Best straight line estimated graphically (Quenouille, 1959).



difference between the underwater equilibrium heart rates of *R. pipiens* and *B. bufo*, examined was there any obvious relationship between body weight and underwater equilibrium heart rate or the proportional reduction in rate at stabilization. There was no apparent difference between the responses of male and female in *R. pipiens* and *R. temporaria*, species in which sex was determined. In an attempt to assess differences between groups of any one species a new population of *R. pipiens* was studied a year after those reported above. There was no significant difference between heart rate of this group, either at the surface or during 1 hr submergence, and the population studied previously. However, arhythmia did not occur so frequently as in the latter and rate of adjustment to underwater equilibrium heart rate was slower ($K = 0.13$) although significantly faster than that observed in *X. laevis* ($K = 0.088$), *B. bufo* ($K = 0.07$) or *R. temporaria* ($K = 0.078$).

Recovery on surfacing was rapid in all species except *B. bufo*. In the latter, complete recovery was not often seen for periods of 30–100 min (Fig. 1). In all species lung ventilation rate was increased (Jones, 1967), but fewer lung ventilating buccal movements were performed per unit time by *B. bufo* than by *R. temporaria* or by *R. pipiens*; *B. bufo* performed between 18–53 (average minimum and maximum) lung "filling or emptying" movements each minute during the first 20 min after surfacing, whereas *R. temporaria* and *R. pipiens* made 68–85 and 73–79 respectively. Recovery was faster in those *B. bufo* which made higher numbers of lung ventilating buccal movements.

(b) *February–March*. Two groups (large and small animals) of *R. temporaria* and *B. bufo* were examined during the breeding season (February–March) of 1 year. Many of the animals were in amplexus and were separated for these experiments. In each group approximately half of the females had laid eggs before the experiments were carried out.

Surface heart rates of both large and small *R. temporaria* were significantly higher than the corresponding groups of toads (Figs. 4 and 5). There was no difference between large and small groups of either species. Bradycardia caused by submersion was not significantly different between species or between large and small animals within one species except during the first 30 min submergence of large *R. temporaria* (Fig. 4a) and large *B. bufo* (Fig. 4b). Exponential decay curves were fitted both for the average values of heart rate and for individuals in order to assess rate of adjustment to underwater equilibrium heart rate. Rate of adjustment was more rapid in the large groups of both species.

Arhythmia occurred in at least 60 per cent of the large animals of both species at some time during development of bradycardia and exceeded 80 per cent in *R. temporaria* after 30 min submergence. Arhythmia occurred much less frequently in the small animals.

(c) *July*. Groups of large and small animals of two species, *B. bufo* and *R. temporaria*, were examined during July of one year. No significant differences existed between either the surface or underwater equilibrium heart rates of the

large and small *R. temporaria* and *B. bufo* (Figs. 4 and 5). Rates of adjustment to underwater equilibrium were not significantly different interspecifically, although large *R. temporaria* adjusted more rapidly than small individuals. At any one time arrhythmia occurred in only 30 per cent of either large *R. temporaria* or large *B. bufo*, although over 50 per cent of small *R. temporaria* exhibited arrhythmia after 30 min submergence.

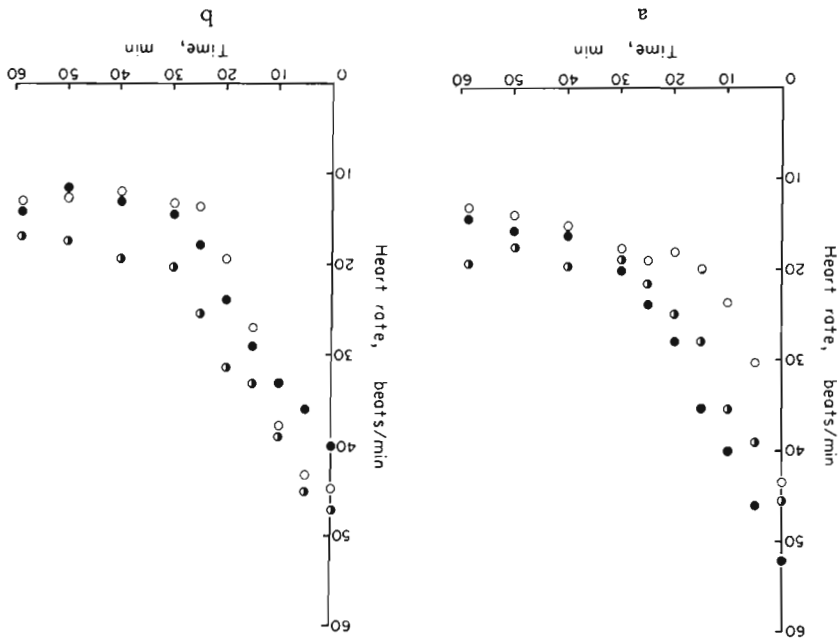


FIG. 4. Seasonal variation in diving bradycardia of large *R. temporaria* and large *B. bufo* during submergence for 1 hr. a. *R. temporaria*, ○, November-December, redrawn from Fig. 1; ●, breeding, 16 experiments, av. wt. = 34.8 ± 1.5 g; ●, summer, 8 experiments, av. wt. = 32.1 ± 1 g. b. *B. bufo*, ○, November-December, redrawn from Fig. 1; ●, breeding, 7 experiments, av. wt. = 41.5 ± 1.2 g; ●, summer, 9 experiments, av. wt. = 43.3 ± 3 g. Submersion at 0 min.

2. Seasonal variation in *R. temporaria* and *B. bufo*

The underwater equilibrium heart rate of summer individuals was higher than that of prehibernation and breeding animals of both species (Figs. 4a, b). Although in large *R. temporaria* (Fig. 4a) the difference between spring and summer was not significant, the trend was confirmed in the small animals where the difference was significant (Fig. 5). Underwater stabilized heart rate of winter *B. bufo* and *R. temporaria* was not significantly different from breeding animals. Winter *R. temporaria* were some 10 g heavier than spring and summer frogs, but a justifiable comparison of underwater equilibrium can probably be made between them since size seems relatively unimportant in this respect.

In both large and small *B. bufo* surface heart rates were lowest in spring, whereas in *R. temporaria* surface heart rates were highest during this period (Figs. 4 and 5). Rate of adjustment to underwater equilibrium heart rate was faster in winter and spring than in summer, apart from small individuals of both species

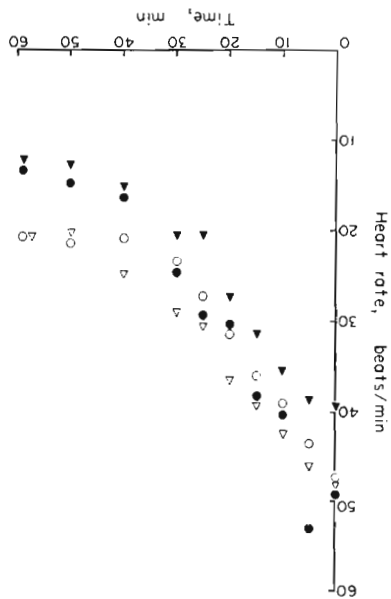


FIG. 5. Seasonal variation in diving bradycardia of small *R. temporaria* and small *B. bufo* during submergence for 1 hr. Submersion at 0 min. ●, *R. temporaria*, breeding, 4 experiments, av. wt. = 23.1 ± 1 g; ○, *R. temporaria*, summer, 12 experiments, av. wt. = 23.1 ± 1 g; ▲, *B. bufo*, breeding, 11 experiments, av. wt. = 24.2 ± 1 g; △, *B. bufo*, summer, 13 experiments, av. wt. = 28 ± 4.5 g.

which showed little variation in this respect. Arrhythmia occurred more frequently in large prehibernation and breeding animals than in summer individuals of both species. In the small animals of both species there was no seasonal difference in the number of animals which developed arrhythmia.

3. Heart rate during attempted hibernation

In order to compare the underwater equilibrium heart rate with that which might occur during winter in the wild, six *R. temporaria*, weighing between 17 and 45 g, were kept at 4–5°C. Heart rates attained during the early part of exposure are shown in Table 1. Differences related to size and sex were not marked. The heart rates were some 4–8 beats/min below those seen during 1 hr submersion at 17°C. Dittmer & Grebe (1959) have shown that up to 25°C, the increase in heart rate with temperature, in the intact frog, follows the relationship

$$Y = 4.8 + 1.1x,$$

where Y = heart rate in beats/min and x = temperature in degrees centigrade. Water temperature during the present experiments was $4-5^{\circ}\text{C}$. If the equation is valid then, at this temperature, heart rate would be between 9 and 10 beats/min.

TABLE 1—HEART RATE (BEATS/MIN) OF *R. temporaria* DURING PROLONGED IMMERSION IN WATER AT 4°C

Sex and wt wt. of frog (g)	Time in water		
	24 hr	70 hr	80 hr
♂ 17.5	10.6	12.0	—
♀ 17.5	8.2	8.0	—
♂ 30	—	—	7.0
♀ 30	—	6.2	—
♂ 45	8.0	—	8.9
♀ 45	8.9	—	7.0

The frogs followed this relationship quite closely (Table 1). Despite low temperature frogs did not become completely quiescent and continued to move around the jars, probably paying visits to the surface. Even after 30 days in the cold frogs still showed intermittent bursts of activity.

4. Variation of diving response after prolonged exposure to laboratory conditions

In order to study the effect on the diving performance of prolonged exposure to laboratory conditions, four experiments were performed using simultaneously toads which had been kept in the laboratory for 9–12 months and recently procured winter animals. The average results are shown in Fig. 6. The two curves are significantly different. Logarithmic transformations showed that the rate of adjustment of heart beat was some 20 per cent slower in laboratory-acclimated animals. Laboratory animals struggled more frequently than winter toads and arrhythmia never developed during submersion. Winter toads usually ceased buccal movements after 15–20 min submergence, whereas laboratory animals continued much longer and in one case persisted throughout the period of submersion. Increase in heart rate following emergence was of the same pattern in both winter and laboratory-acclimated animals. There was an initial rapid increase of about 10 beats/min at the start of respiratory movements. Further increases in rate, in intervals between the recording periods, were usually smaller than this and were approximately the same for both winter and laboratory-acclimated animals. Consequently the predictive rate was achieved more rapidly in the laboratory-acclimated toads since bradycardia was not so marked.

DISCUSSION

A seasonal variation in the response of heart rate to submergence exists in *R. temporaria* and *B. bufo*; changes in both rate of development of diving

bradycardia and underwater stabilized heart rate occur which, although statistically significant in some cases, are not so marked as to make seasonal characterization of animals possible. The greatest variation in bradycardia is between prehibernation and summer with breeding animals being somewhat intermediate in the case of *R. temporaria* but resembling prehibernation animals in *B. bufo*. The multiplicity

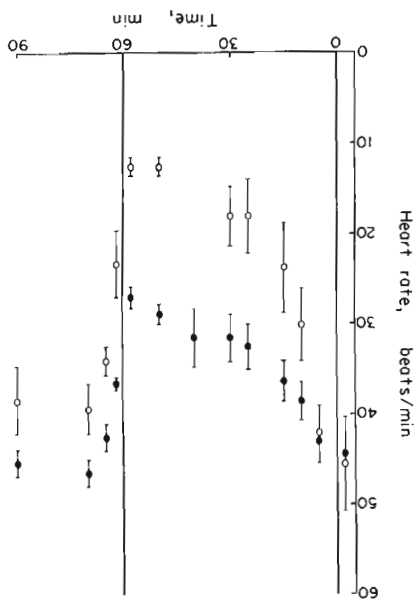


FIG. 6. Bradycardia caused by submergence and recovery on surfacing in laboratory-acclimated and winter *B. bufo*. Two toads, one from each group, were examined simultaneously. Errors on mean indicated. ●, Laboratory acclimated, four toads, av. wt. 38.4 ± 2.8 g.; ○, November-December, four toads, av. wt. 41.5 ± 3.1 g. Submersion and emersion 0 and 60 min respectively.

of factors which have been observed to show seasonal variations in amphibia attains peaks usually associated with the breeding season or winter torpor and, on this basis, variations in diving bradycardia would appear to be independent of many of these.

With the present information any explanation of seasonal variation in diving bradycardia must be incomplete. There is no information regarding how long the underwater equilibrium heart rate can be maintained. It is tempting to suggest that the heart is in a steady state with its surroundings and that activity can continue, at this level, for long periods. If this is so then it seems safe to assume that metabolism is "basal" during the last half hour of submergence. Heart rate certainly gives an indication of the level of metabolic activity. Reduction in heart rate, oxygen consumption and heat production occurs simultaneously during

submergence (Leivestad, 1960; Jones, 1967), and in *R. temporaria* and *B. bufo*, which have been acclimated to laboratory conditions, bradycardia during the last half of 1 hr submergence in agitated water bears a relationship to cutaneous oxygen consumption per unit weight (Jones, 1967). Assuming that a relationship of this type is valid in regard to the present experiments, then variations in underwater stabilized heart rate are best explained in these terms. On this premise, basal oxygen uptake in summer animals must be higher than that of breeding or pre-hibernating frogs and toads. Consequently winter animals are capable of withstanding a reduction in oxygen metabolism to a lower level than in summer. This seems advantageous in that the metabolic reserves of winter animals are probably depleted at a slower rate than in summer frogs and toads, thereby ensuring survival if submergence is prolonged, particularly in the case of *R. temporaria*, which may hibernate under water. This presupposes the existence of a specific "winter condition" as opposed to that in summer animals. In fact *R. pipiens* only hibernates in the laboratory at 0°C between October and April which lends support to this conclusion.

There are no data on seasonal variations in cutaneous oxygen exchange, in the absence of pulmonary ventilation, for either *R. temporaria* or *B. bufo*. Basal oxygen uptake of *R. pipiens* (Fromm & Johnson, 1955) is highest during the breeding season. However, the condition of the animals before the experiments is important in determining basal oxygen consumption. Refrigeration at 4°C causes a significant reduction in oxygen uptake of about 25 per cent compared with frogs kept at 22-28°C during autumn, winter and spring (Fromm & Johnson, 1955). *R. temporaria* particularly and *B. bufo* breed during February-March when environmental temperatures are low and for this reason are not comparable to the spring population of *R. pipiens* maintained at 22-28°C, but to a refrigerated group which had a basal oxygen uptake intermediate between that of winter and summer animals (Fromm & Johnson, 1955).

Simultaneous submersion of winter and laboratory-acclimated toads of the same weight confirmed that the difference in degree of bradycardia is due to the conditions which the animal experiences before the experiment. Laboratory-acclimated toads resemble summer animals in both heart rate and behavioural response to submersion. Furthermore, it appears that the difference between laboratory and winter animals is extrinsic rather than intrinsic. Holzapfel (1937) suggested that hibernation was one phase of the seasonal cycle which is intrinsic in both *R. pipiens* and *R. catesbeiana*. Providing it is accepted that enhanced diving bradycardia is an indication of an hibernating condition, then, since photoperiod was the same for both winter and laboratory-acclimated toads, temperature and possible shortage of food may be the environmental factors involved in onset of hibernation.

Adjustment of heart rate to underwater equilibrium rate occurs faster in winter than summer frogs and toads. Variations in oxygen uptake may be responsible but the occurrence of pronounced arrhythmia in winter but not summer animals indicates that other agencies may be involved. Arrhythmia does not occur in

vagatomized frogs during development of bradycardia (Jones & Shelton, 1964) and appears to be caused by inhibitory vagal activity. This observation supports the proposal that reduction in heart rate during submergence can be achieved by different means, although hypoxia is probably the driving force in both cases. Hypoxia may have a direct effect on enzymatic or cellular systems within the pacemaker region, which results in decreased heart rate, or alternatively hypoxia may trigger nervous inhibition of the heart which causes arrhythmia. The amount of interaction between these two factors is not known but, based on the evidence of numbers of animals showing arrhythmia, nervous inhibition predominates during winter whereas the "direct effect" predominates in summer animals. In fact activity in the parasympathetic nervous system appears to be seasonal, for Iriuchijima (1959) recorded parasympathetic impulses from the cardiac nerve of the toad only during November and February. Breeding animals are somewhat intermediate, neither the direct affect of hypoxia nor nervous inhibition of the heart is dominant. Many develop arrhythmia but after a longer period of submergence than winter animals. Heart rate is often low when this occurs and its effect on rate of adjustment to underwater equilibrium is consequently not so marked as in winter frogs and toads.

Underwater heart rate during attempted hibernation may be governed, at least in part, directly by temperature. *R. temporaria* does not become completely torpid during hibernation under water and probably pays visits to the surface on a warm day (Mertens, 1947); on submersion nervous inhibition causes a rapid lowering of heart rate and when the frog has cooled to the ambient temperature nervous control may be superseded by temperature affecting the heart directly. By this means a rapid adjustment of heart rate, and probably metabolism, to the hibernating condition would be achieved thereby conserving vital resources. Holzapfel (1937) found that vagus tone was practically absent in hibernating *R. pipiens* (at 0°C), which suggests that the hibernating heart rate, which is low in *R. temporaria* of a wide size range, may be entirely independent of nervous control.

Little correlation between habitat and response of the heart to submersion exists in animals examined before a period of natural quiescence. Apart from *X. laevis*, the species which normally spend this period on land and those which may pass this period in water show a similar response, with respect to the final stabilized heart rate, although the development of full bradycardia is much faster in *R. pipiens* than in the others. Arrhythmia occurred in nearly all *R. pipiens* during the first 15 min of submersion and in some persisted throughout. This pattern was not consistently seen in the other species and points to greater vagal inhibition of the heart in *R. pipiens*. The significance of rapid adjustment in *R. pipiens* is obscure. The lowest underwater equilibrium heart rate, 2-3 beats/min in one animal, was found in *X. laevis*. This seems to be a reflection of a difference in metabolic activity between *X. laevis* and the other species rather than a direct environmental response. Surface heart rate of *X. laevis* is also below that of individuals of other species of the same size and weight; and recorded values of oxygen uptake per unit weight both before and during submergence (Charles,

1931; Jones, 1967) are usually lower than those quoted for other anurans (Fromm & Johnson, 1955; Jones, 1967). During the breeding season and summer months there is no difference between the responses to submergence of the predominantly terrestrial *B. bufo* and *R. temporaria*, which is more aquatic in its behaviour.

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