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THE REFLEX CONTROL OF HEART RATE AND CARDIAC OUTPUT IN THE RAINBOW TROUT: INTERACTIVE INFLUENCES OF HYPOXIA, HAEMORRHAGE, AND SYSTEMIC VASOMOTOR TONE

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SUMMARY

In cannulated trout there was no cholinergic vagal tone as revealed by atropine blockade during normal heart rates. Reductions in heart rate occasionally occurred under normoxia without apparent external stimuli ('spontaneous' bradycardia) and always occurred under environmental hypoxia (hypoxic bradycardia) due to the imposition of significant vagal tone. Direct measurements of cardiac output ($\dot{Q}$) during these bradycardias showed that increases in cardiac stroke volume compensated for the falls in heart rate so that total $\dot{Q}$ remained unchanged or increased slightly. Sudden experimental reductions in arterial blood pressure via blockade of systemic vasomotor tone with yohimbine or via haemorrhage had no effect on heart rate during normal rates, but caused cardioacceleration during both types of bradycardia. These increases in heart rate never exceeded the point of zero vagal tone (normal heart rate) and were largely or wholly due to reductions in endogenous vagal tone. These cardioaccelerations were temporary; spontaneous bradycardia could re-occur at any time, while hypoxic bradycardia always re-occurred if the hypoxic stimulus were maintained. The results are interpreted in terms of a central interaction between the baroreceptor and chemoreceptor reflexes.

INTRODUCTION

It has long been known that environmental hypoxia can elicit a reduction in heart rate in teleosts (e.g. Randall & Shelton, 1963) as can a rise in arterial blood pressure (e.g. Mott, 1951). A variety of non-specific 'stresses' can also induce bradycardia (e.g. Randall, 1968). The efferent pathways for all these reflexes are the parasympathetic fibres of the cardiac branch of the vagus nerve. Hypoxia receptors (chemoreceptors) have recently been localized to the first pair of branchial arches (Daxboeck & Holeton, 1978; Smith & Jones, 1978) while baroreceptors apparently occur on all four gill arches (Ristori, 1970; Ristori & Dessaux, 1970). Parts of these arches are considered the phylogenetic antecedents of the peripheral reflexogenic areas of the mammalian circulation, the carotid and aortic regions (Heymans & Neil, 1958).

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In mammals, complex interactive effects of peripheral chemoreceptor and baroreceptor stimulation on cardiovascular function are well documented, though not completely understood (Heymans & Neil, 1958; Burton, 1972; Berne & Levy, 1972). Recent work indicates that such interactions may result from direct inputs of both receptor types to the medullary vasomotor centre, from interconnexions between the respiratory and vasomotor centres, and from secondary systemic reflexes (Öberg, 1976). In teleost fish, both baroreceptor reflexes (Mott, 1951; Randall & Stevens, 1967; Ristori, 1970; Ristori & Dessaux, 1970; Stevens, Bennion, Randall & Shelton, 1972; Helgason & Nilsson, 1973; Wood & Shelton, 1980) and chemoreceptor reflexes (Randall & Shelton, 1963; Holeton & Randall, 1967*a*, *b*; Randall & Smith, 1967; Daxboeck & Holeton, 1978; Smith & Jones, 1978) have been well documented, but there is no information on possible interactions between the two. The primary aim of the present study on the rainbow trout was to investigate whether such interactions in fact occur, employing changes in heart rate as an index of the cardiovascular reflexes. The previous baroreceptor studies have concentrated only on the results of loading the pressoreceptors. Under such loading, interactive effects would be difficult to detect, because both pressor and hypoxic stimuli have similar inhibitory influences on heart rate. Therefore the present study examined the effects of unloading the baroreceptors during both normoxic and hypoxic conditions. A second objective was to directly measure the influence of the bradycardial response on total cardiac output.

MATERIALS AND METHODS

The experiments were performed on 40 rainbow trout (*Salmo gairdneri*) as part of a general study on the effects of autonomically active drugs on cardiovascular performance, some of which has already been described (Wood & Shelton, 1980). The maintenance, operative, experimental, and recording conditions are detailed in that report. Thirty-five of the animals were fitted with dorsal aortic, subintestinal vein, and either caudal artery or caudal vein catheters. The dorsal aortic cannula was used for recording arterial blood pressure (P_a) and instantaneous heart rate, the caudal artery and vein cannulae for experimental haemorrhage, and the subintestinal vein cannula for injection of drugs. The other five fish each received a ventral aortic flow probe for the direct measurement of cardiac output ($\dot{Q}$) in addition to the combination of catheters. Cardiac stroke volume was calculated from simultaneous measurements of mean ventral aortic flow and instantaneous heart rate. Drugs employed were l-noradrenaline bitartrate, l-adrenaline bitartrate, yohimbine hydrochloride, acetylcholine chloride, and atropine sulphate (all Sigma). Other more specific experimental details are given at appropriate points in the text.

Most fish were exposed to a sequence of drugs and treatments, but each experimental run was separated by at least 4 h. After pharmacological blockade (yohimbine, atropine), further tests were not carried out for at least 48 h to ensure that the effects of the blockade had totally disappeared. The efficacy of blockade was periodically checked with appropriate agonists (acetylcholine for muscarinic cholinergic blockade, adrenaline or noradrenaline for α-adrenergic blockade; Wood & Shelton, 1980). Dorsal aortic blood pressures are reported as area means – i.e. 1/3 (1 systolic + 2 diastolic) (Burton, 1972). Throughout the study, each animal was used as its own

control. Results have been expressed as $\bar{x} \pm 1$ S.E. (N), where N represents the number of experimental runs. The significance of differences before and after a particular experimental procedure were tested by means of the Student's paired two-tailed t test. Except in the acetylcholine and $\dot{Q}$ trials, at least five trout contributed data to each experimental treatment group.

RESULTS

(I) *Spontaneous bradycardia*

Under normoxic conditions (P_{I,O_2} = 140–155 torr), the present trout normally maintained mean P_a's of 25–45 cm H_2O and regular heart rates of 60–110/min with generally little variation of the instantaneous beat to beat frequency as recorded by the ratemeter. However nearly all the animals exhibited occasional periods (30 s to 20 min) of reduced heart rate (30–70/min) usually accompanied by an irregularity of the instantaneous frequency (Fig. 1, Table 1). Arterial pulse pressure was increased and mean P_a was sometimes slightly elevated (Table 1). Direct measurements of $\dot{Q}$ were obtained in two trout (one run each) during the bradycardia. These indicated that the fall in heart rate was accompanied by an almost proportional increase in cardiac stroke volume so that total $\dot{Q}$ was more or less constant. The event was not associated with any apparent external stress. For convenience, it is termed 'spontaneous' bradycardia, although we recognize that unknown external stimuli may have been the proximate cause.

The muscarinic cholinergic antagonist atropine (100 nmole/100 g) immediately and permanently (4–6 h) reversed the spontaneous bradycardia back to normal heart rate (Table 1, Fig. 1A). Atropine had no effect on cardiac frequency during normal heart rate (Table 1). These results indicate that spontaneous bradycardia is a classical 'vagal bradycardia' mediated through muscarinic cholinergic receptors on the heart (cf. Randall, 1968). During both normal and spontaneously reduced heart rates, atropine also caused a significant fall in P_a (Table 1, Fig. 1A) which persisted for only 15–30 min. Measurements of $\dot{Q}$ under normal heart rates indicated that the latter was at least in part associated with a depressant effect of the drug on cardiac stroke volume (C. M. Wood & G. Shelton, in preparation).

The α-adrenergic antagonist yohimbine (100 nmole/100 g), when administered during spontaneous bradycardia. had a superficially similar effect to that of atropine. The heart rate returned to normal and P_a fell markedly (Fig. 1B, Table 1), the latter due to blockade of endogenous α-adrenergic tone in the systemic vasculature (Wood & Shelton, 1980). Like atropine, yohimbine similarly reduced Pa during normal heart rates but had no effect on cardiac frequency (Table 1). At least during normal heart rates, yohimbine had a negligible effect on $\dot{Q}$ and cardiac stroke volume (Wood & Shelton, 1980). However, whereas the reversal of bradycardia by atropine was immediate and preceded the drop in P_a (Fig. 1A), the reversal by yohimbine was delayed and paralleled or followed the fall in Pa (Fig. 1B). Furthermore, the depressor effect of yohimbine was extremely long-lasting (6–12 h), but spontaneous bradycardia could re-occur at any time, sometimes within minutes. These results suggested that the fall in systemic vascular resistance (R_s) mediated by yohimbine (Wood & Shelton, 1975; Wood, 1976; Wood & Shelton, 1980) was itself responsible for the temporary reversal of spontaneous bradycardia. This could occur through the drop in R_s either directly

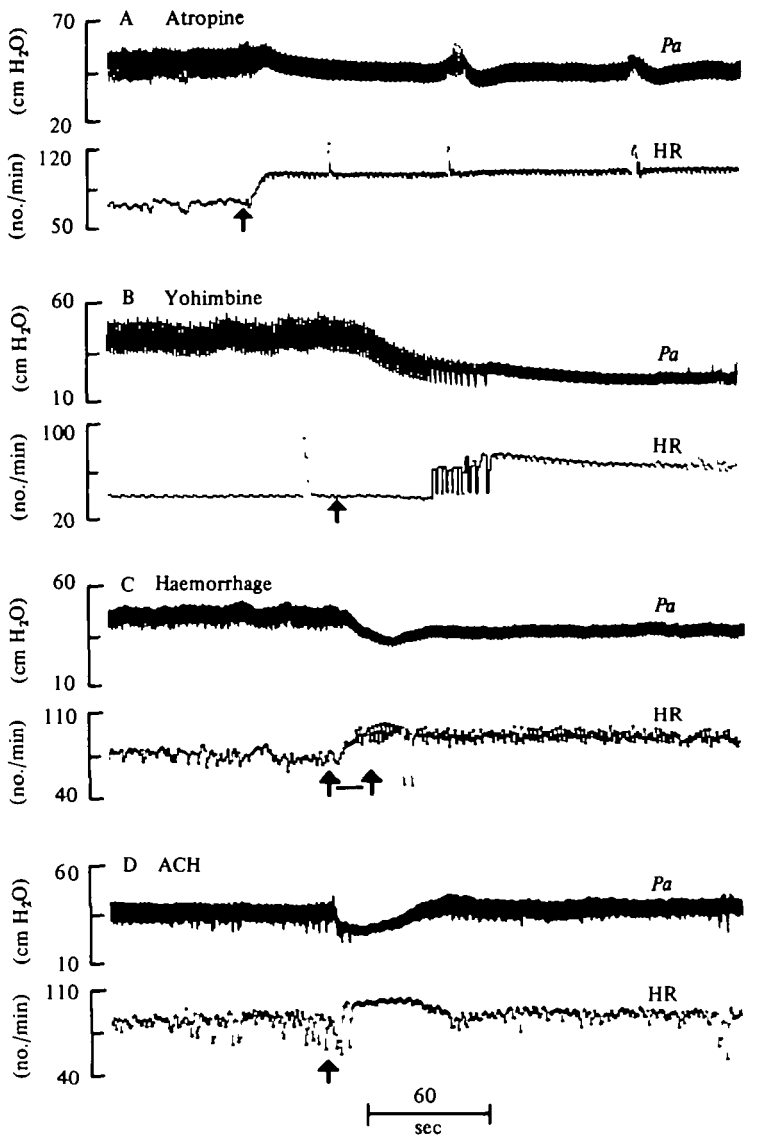

Fig. 1. Original records showing the effects of four different treatments (at arrows) on instantaneous heart rate (HR) and dorsal aortic blood pressure (P_a) during periods of spontaneous bradycardia in four different fish. (A) atropine (100 nmole/100 g). (B) yohimbine (100 nmole/100 g). (C) haemorrhage (0·5 % body weight) from caudal vein. (D) acetylcholine (320 pmole/100 g).

(via neural feedback from, say, length receptors in the smooth muscle of systemic resistance vessels) or indirectly (via the consequent fall in P_a acting on baroreceptors) overriding the bradycardial mechanism at a central level. However, the possibility remained that yohimbine could be non-specifically blocking cardiac muscarinic receptors and therefore reversing spontaneous bradycardia by essentially the same mechanism as atropine. This possibility was eliminated by the results of numerous experiments in which this dose of yohimbine had no effect on the cardiac dos-

Table 1. *The effects of atropine* (100 *nmole*/100 *g*), *yohimbine* (100 *nmole*/100 *g*), *and haemorrhage* (0·5% *body weight*) *on heart rate* (*HR*) *and average dorsal aortic blood pressure* (P_a) *during periods of normal heart rate and spontaneous bradycardia* (*means* ± 1 S.E.)

Treatment	Before treatment		After treatment	
	P_a (cm H_2O)	HR (beats/min)	P_a (cm H_2O)	(HR (beats/min)
		Normal heart rate		
Atropine (N = 14)	32·7 ± 1·8	69·4 ± 3·5	26·7 ± 1·2 P < 0·001	69·2 ± 3·3 P = n.s.
Yohimbine (N = 10)	33·6 ± 2·1	78·5 ± 3·8	21·7 ± 1·4 P < 0·001	78·0 ± 4·0 P = n.s.
Haemorrhage (N = 6)	37·8 ± 3·4	73·3 ± 3·2	25·8 ± 2·2 P < 0·001	73·4 ± 3·1 P = n.s.
		Spontaneous bradycardia		
Atropine (N = 6)	40·1 ± 2·6	58·9 ± 5·3	33·0 ± 3·5 P < 0·01	85·6 ± 5·9 P < 0·01
Yohimbine (N = 6)	30·6 ± 3·3	44·6 ± 4·4	20·1 ± 1·8 P < 0·02	75·3 ± 3·8 P < 0·01
Haemorrhage (N = 12)	43·1 ± 1·8	56·0 ± 5·0	33·1 ± 2·0 P < 0·001	74·6 ± 4·1 P < 0·001

P = significance with respect to corresponding value before treatment.

response curve (negative chronotropism) to injected acetylcholine (C. M. Wood & G. Shelton, in preparation).

Experimental haemorrhage was employed to test whether the reversal of spontaneous bradycardia by yohimbine was directly due to the decreases in R_s or indirectly mediated by the resulting fall in blood pressure. Haemorrhage should reduce P_a without directly lowering R_s. Indeed, if anything, the procedure would be expected to increase R_s by reduction of passive distention (Wood & Shelton, 1975). Animals were bled by 0·5% body weight (approximately 10% blood volume) over 15–30 s from either the caudal artery or caudal vein catheter while P_a and heart rate were recorded from the dorsal aortic catheter. The rate of decline of P_a, but not the final size of the decrease, was somewhat greater with bleeding from the arterial than from the venous site, but results were very similar with the two procedures and are combined in subsequent analyses.

Haemorrhage caused virtually identical effects to those produced by yohimbine. Spontaneous bradycardia was reversed with a time course following or paralleling the fall in P_a (Fig. 1 C, Table 1). Again the effect was by no means permanent, and bradycardia could be re-imposed at any time. Haemorrhage during normal heart rates caused a comparable fall in P_a but had no effect on cardiac frequency (Table 1). Re-infusion of the blood after 3–5 min always caused a pressor effect in P_a (an increase of 15–40 cm H_2O) and accompanying bradycardia of variable duration in trout that originally had either normal or spontaneously reduced heart rates. Thus it is not the fall in R_s itself caused by yohimbine, but rather the resulting fall in pressure that is important in overriding spontaneous bradycardia. Further support for this concept was gained by reducing blood pressure in a totally different manner in three fish. Low doses of acetylcholine (up to 1 nmole/100 g) decrease blood pressures throughout the cardiovascular system by muscarinic inhibition of cardiac stroke volume with no

effect on the systemic vasculature (Wood, 1977; C. M. Wood & G. Shelton, in preparation). Such doses either had no effect or an inhibitory effect on cardiac frequency during normal heart rates, but during spontaneous bradycardia caused a definite increase (Fig. 1 D). The bradycardia was usually re-established as the depressor effect subsided. All the evidence therefore points to sensory input from baroceptors interacting at a central level with the mechanism of bradycardia and releasing vagal tone.

Measurements of $\dot{Q}$ during haemorrhage were obtained in two fish, but only during normal heart rates. Haemorrhage caused a rapid decline in cardiac stroke volumes (−42%, −32%) which remained stable at these reduced levels over the next 3–5 min. As heart rates did not change, these reductions were directly reflected in $\dot{Q}$. Reinfusion of the blood at the end of this period caused a temporary bradycardia (15–30 s) with sustained increases in stroke volume and $\dot{Q}$ above control levels (+37%, +38%) followed by a gradual return to normal after 10–15 min. In order to follow the time course of P_a correction after unreplaced haemorrhage, the blood was not immediately re-infused in some experiments. In these cases, the fall in P_a was gradually and completely corrected over 15–40 min.

II. *Hypoxic bradycardia*

A parallel series of experiments was performed during hypoxic bradycardia to test whether a similar effect of blood pressure on heart rate occurred during a bradycardia of chemoreceptor origin. Environmental hypoxia was induced by simply stopping the water flow to the fish chambers and allowing the animal to deplete the available oxygen. The treatment was controlled not by the change in water P_{O_2} but by the change in heart rate. The chamber was closed until cardiac frequency had declined by 25–50% of the normal level, a period of 20–45 min, depending on the size of the fish relative to the size of the chamber. Measurements of P_{I,O_2} and P_{I,CO_2} were performed in some trials. P_{I,O_2} typically declined from 140–155 torr to 50–65 torr by the end of the hypoxic period, while P_{I,CO_2} increased from about 0·5 to 1·0 torr.

A highly significant bradycardia occurred during hypoxia in all treatment groups (Table 2). P_a rose in magnitude and pulse amplitude in most (but not all) fish (e.g. Fig. 2 A), and the overall change was significant in most groups (Table 2). It is possible that the rise in P_a could have contributed to the bradycardia via the baroreceptor reflex in some of the fish, but the extent of heart slowing was much greater than that normally associated with pressor events of this magnitude in non-hypoxic fish. The rise in P_a in one treatment group (control) was more than twice as large as in the others, yet the bradycardia was of similar magnitude (Table 2). Furthermore the hypoxic bradycardia developed normally after the pressor response had been eliminated by yohimbine (see below). A specific effect of hypoxia mediated via chemoreceptors was therefore indicated. In control fish (subjected only to hypoxia), flushing the chamber with normoxic water at the end of the hypoxic period caused an immediate increase back to the original rate often accompanied by a further small rise in P_a (Table 2). By 30 s after the end of hypoxia, P_a had stabilized at a level significantly lower than the hypoxic value but higher than the original (Table 2) and gradually returned to normal over the following 30–60 min.

The administration of atropine (100 nmole/100 g) during hypoxic bradycardia immediately returned heart rate to the original level despite the continuation of hypox-

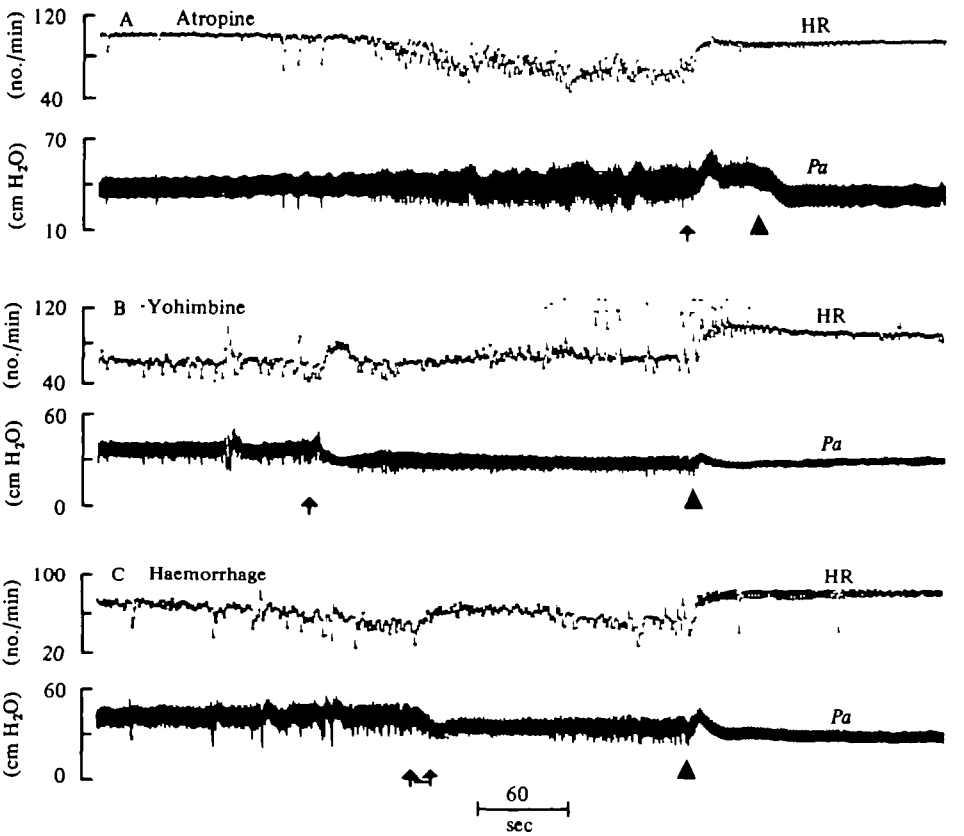

Fig. 2. Original records showing the effects of three different treatments (at arrows) on instantaneous heart rate (HR) and dorsal aortic blood pressure (P_a) during periods of environmental hypoxia in three different fish. (A) Atropine (100 nmole/100 g). (B) yohimbine (100 nmole/100 g). (C) Haemorrhage (0·5 % body weight) from caudal vein. Hypoxia was induced by stopping the water flow to the fish chambers prior to the start of the period shown. Note the development of the hypoxic bradycardia in (A) while it was already partially developed in (B) and (C) prior to the start of the record shown. At ▲ the chambers were flushed with normoxic water.

(Fig. 2A, Table 2). P_a did not change significantly (Table 2). Flushing the chamber with normoxic water caused no further change in heart rate, but P_a dropped to a level significantly below the original (Fig. 2A, Table 2). Subsequent imposition of hypoxia had no effect on heart rate. Thus the hypoxic bradycardia was also of classical vagal origin (cf. Randall & Smith, 1967).

Yohimbine (100 nmole/100 g) injected during hypoxic bradycardia caused a dramatic fall in P_a (Fig. 2B, Table 2) similar to that seen during normal heart rate or spontaneous bradycardia (Fig. 1B, Table 1). The hypoxic bradycardia was reversed to a variable but significant extent during or following the fall in P_a (Fig. 2B). The maximum rate reached during this cardioacceleration was never greater than the original in any run, and, on average, remained significantly lower than both the original and post-hypoxia values (Fig. 2B, Table 2). As hypoxia was continued, the heart rate eventually returned to the pre-yohimbine level of bradycardia, though the time course of the return was extremely variable from fish to fish. Fig. 2B shows an

Table 2. *The effects of environmental hypoxia and its alleviation on heart rate (HR) and average dorsal aortic blood pressure (P_a), and the effects of atropine (100 nmole/100 g), yohimbine (100 nmole/100 g), and haemorrhage (0·5% body weight) on these responses (means ± 1 S.E.)*

	Original		Hypoxia		After treatment		After hypoxia	
Treatment	P_a (cm H_2O)	HR (no./min)	P_a (cm H_2O)	HR (no./min)	P_a (cm H_2O)	HR (no./min)	P_a (cm H_2O)	HR (no./min)
Control ($N = 9$)	33·5 ± 2·5	86·1 ± 3·5	42·6 ± 4·0 $P < 0.01$	52·8 ± 4·2 $P < 0.001$	—	—	38·5 ± 3·8 $P < 0.05$ $P_0 < 0.05$	86·1 ± 3·0 $P < 0.001$ P_0 = n.s.
Atropine ($N = 5$)	34·4 ± 2·2	85·8 ± 6·3	36·6 ± 3·2 P = n.s.	55·0 ± 5·5 $P < 0.001$	39·6 ± 3·0 P = n.s.	89·6 ± 5·7 $P < 0.001$	30·2 ± 2·5 $P < 0.02$ $P_0 < 0.01$	87·7 ± 6·9 P = n.s. P_0 = n.s.
Yohimbine ($N = 7$)	36·4 ± 3·0	92·1 ± 3·8	40·7 ± 4·1 $P < 0.05$	61·1 ± 5·7 $P < 0.01$	26·6 ± 3·7 $P < 0.01$	74·9 ± 6·6 $P < 0.05$	26·9 ± 3·6 P = n.s. $P_0 < 0.02$	94·4 ± 4·5 $P < 0.02$ P_0 = n.s.
Haemorrhage ($N = 10$)	39·0 ± 2·4	88·3 ± 4·2	41·3 ± 2·6 $P < 0.02$	56·3 ± 2·5 $P < 0.001$	29·8 ± 2·7 $P < 0.001$	80·5 ± 3·5 $P < 0.001$	32·4 ± 3·0 P = n.s. $P_0 < 0.01$	92·3 ± 4·4 $P < 0.001$ P_0 = n.s.

P = significance with respect to corresponding value in the preceding category.

P_0 = significance with respect to corresponding value in the 'Original' category.

Notes: (1) Environmental hypoxia was induced by stopping the water flow to the fish chamber. (2) 'Original' values were determined prior to hypoxia. (3) 'After Treatment' values were determined at the point after treatment where heart rate was maximal. (4) 'After Hypoxia' values were determined 30 s after flushing the chamber with normoxic water.

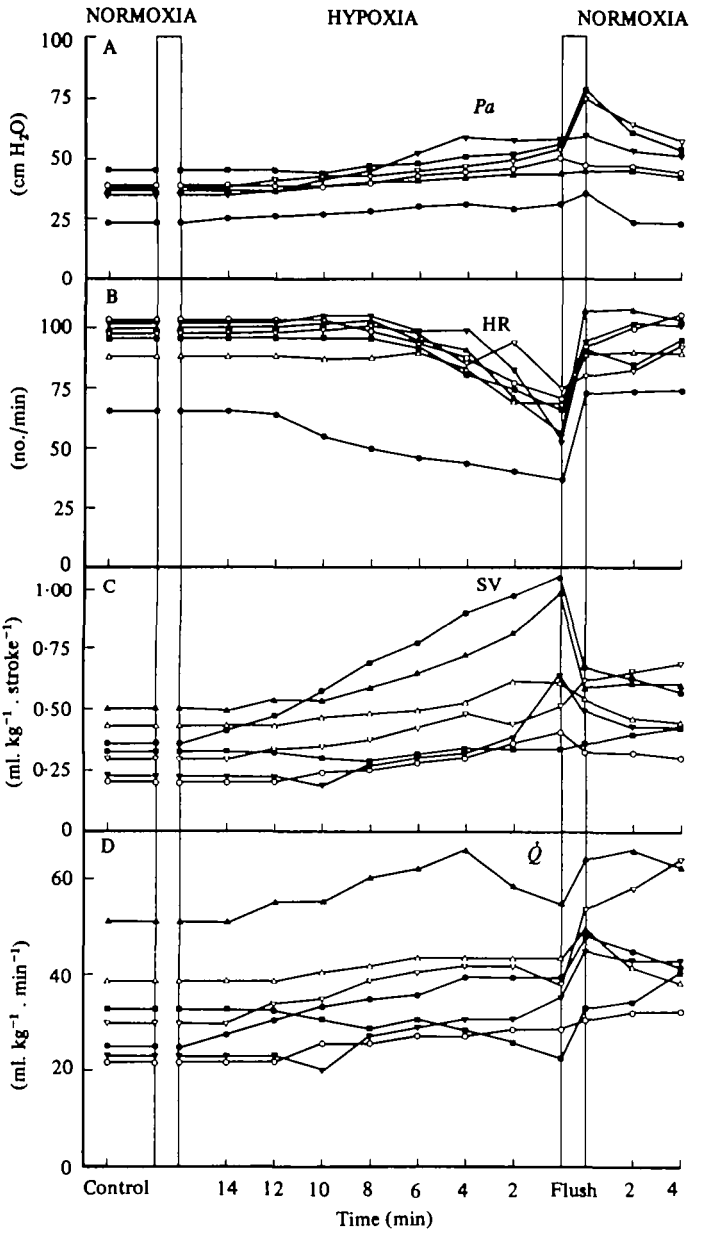

Fig. 3. Changes in (A) mean dorsal aortic blood pressure (P_a); (B) heart rate (HR); (C) cardiac stroke volume (SV); and (D) total cardiac output ($\dot{Q}$) during the last 16 min of environmental hypoxia and after return to normoxia in seven runs on three fish. Control values were taken prior to hypoxia. Hypoxia was induced by stopping the water flow to the fish chambers at some time (4–29 min. in different experiments) prior to the start of the 16 min hypoxic period shown here. Normoxia was reintroduced by opening the flow to the chambers (flush). ▼, ▽, ■, △ = 718 g; ○, ▲ = 681 g; ● = 148 g. P_a records were not obtained in △ due to cannula failure.

example where the yohimbine cardioacceleration was extremely short-lived. Re-imposition of normoxia caused an immediate return to normal heart rate as in control fish, but P_a remained significantly depressed (Table 2, Fig. 2B). Subsequent imposition of hypoxia provoked the normal bradycardial response with essentially no change in P_a.

The effects of haemorrhage (0·5% body weight) on heart rate during hypoxia were again very similar to those of yohimbine. Bleeding caused a reversal of bradycardia paralleling or following the fall in P_a. This cardioacceleration was of variable extent but never exceeded the original heart rate; it was always eventually overcome by the continuation of hypoxia (Fig. 2C, Table 2). As with yohimbine, the average maximum rate after haemorrhage remained significantly lower than both the original and post-hypoxia values. Flushing with normoxic water produced an immediate recovery to the original rate but P_a remained significantly depressed for 15–40 min or until the blood was re-infused (Fig. 2C, Table 2). Subsequent responses to hypoxia were completely normal.

Thus a fall in R_s interacts with hypoxic bradycardia in a similar fashion to its interaction with spontaneous bradycardia. It is the resulting fall in blood pressure, rather than the drop in R_s itself, which is the proximate stimulus. Apparently the change in baroreceptor stimulation caused by the depressor effect is sufficient to overcome, at least temporarily, chemoreceptor drive and thus alleviate bradycardia. However as the hypoxia becomes more severe (and possibly the sensitivity of the baroreceptors is adjusted) the chemoreceptor drive tending to reduce heart rate again becomes dominant and the bradycardia is re-imposed.

Direct measurements of $\dot{Q}$ during hypoxic bradycardia were obtained in seven runs on three fish (Fig. 3). In order to prevent unnecessary struggling and possible displacement of the flow probe, the duration and degree of hypoxia (P_{I,O_2} = 60–80 torr) were reduced as compared to the experiments described above. Nevertheless changes in P_a (Fig. 3A) and heart rate (Fig. 3B) followed the same trends as in the other groups (Table 2). The results of these seven runs clearly showed that the fall in heart rate (from 92·9 ± 4·9 to 62·0 ± 4·9/min; $P < 0{\cdot}001$, Fig. 3B) was more than compensated for by an approximate doubling of cardiac stroke volume (from 0·340 ± 0·043 to 0·645 ml/kg.stroke, $P < 0{\cdot}02$; Fig. 3C) during hypoxia. On average, $\dot{Q}$ actually increased slightly (from 31·23 ± 4·07 to 37·50 ± 3·94 ml/kg.min^{-1}, $0{\cdot}05 < P < 0{\cdot}1$; Fig. 3D). Flushing the chamber with normoxic water caused an immediate further rise in $\dot{Q}$ (to 45·60 ± 4·25 ml/kg.min, $P < 0{\cdot}001$; Fig. 3D) because the rapid increase in heart rate (Fig. 3B) was not accompanied by a compensatory reduction in stroke volume. Indeed stroke volume actually increased further in two runs (Fig. 3D) and on average (0·517 ± 0·048 ml/kg.stroke) remained considerably above the original normoxic value ($P < 0{\cdot}01$). Cardiac output and stroke volume thereafter gradually returned to normal over the following 30–60 min.

DISCUSSION

The present results clearly show that interaction occurs between baroreceptor and chemoreceptor reflexes in the rainbow trout. In particular, unloading the baroreceptors by any procedure which reduces blood pressure can temporarily override the chemo-

receptor drive that tends to reduce heart rate during environmental hypoxia. Indeed the same sort of interaction seems to occur even when the bradycardial stimulus is not apparently of chemoreceptor origin ('spontaneous bradycardia'). It could be argued that such sudden vasodepression could directly change chemoreceptor stimulation by altering blood flow or oxygen delivery to the receptors, and thereby contribute to the interaction. However, this is unlikely if the oxygen receptors concerned are external rather than arterial as Randall & Smith (1967) and Daxboeck & Holeton (1978) have suggested. In any case, vasodepression should cause a reduction in blood flow and oxygen delivery and therefore an intensification rather than an alleviation of chemoreceptor drive. The major extra-cardiac influence on arterial blood pressure in trout is normally systemic vasomotor tone (Wood & Shelton, 1980), but the results of the yohimbine, haemorrhage and acetylcholine experiments indicate that the interactive mechanism does not rely on sensory feedback supplying information on the diameter of the systemic resistance vessels. Rather there appears to be a central interaction between the chemoreceptor and baroreceptor reflexes which involves direct input from the respective receptor types.

In mammals, a central mechanism also seems to be an important component of interaction between baroreceptor and chemoreceptor reflexes. However, beyond this, it is impossible at present to draw other analogies because no clear concensus has yet emerged in the mammalian literature. The situation is complicated both by conflicting reports (eg. Heistad, Abboud, Mark & Schmid, 1974; Mancia, 1975; Wennergren, Little & Öberg, 1976) and by the presence of opposing primary and secondary reflex mechanisms (Berne & Levy, 1972).

In the present experiments there was normally no resting vagal tone in the trout for atropine had no effect on heart rate. Vasodepressor procedures (yohimbine, haemorrhage, acetylcholine) also did not cause cardioacceleration during normal heart rates though they were highly effective in this regard during spontaneous or hypoxia induced bradycardia. The heart rates resulting from such depressor stimuli were never greater than normal resting values, indicating that an apparent upper limit for the cardioacceleratory effect of baroreceptor unloading is set by the point of zero vagal tone. This implies, though does not prove, that the cardioacceleratory mechanism involved is simply one of reducing vagal tone, for sympathetic stimulation can increase the heart rate above the point of zero vagal tone (Wood, Pieprzak & Trott, 1980).

The finding of zero vagal tone under normal heart rates is in agreement with previous studies on trout (Randall & Smith, 1967; Stevens & Randall, 1967; Priede, 1974). However, recently we have shown that this is not the case in truly resting animals, where blockade of vagal tone with atropine raises the heart rate by about 30% at a comparable temperature (Wood *et al.* 1980). The lack of vagal tone in the present and previous studies probably reflected experimental stress: confinement, surgery, and blood loss (Wood *et al.* 1980). Therefore one might expect to see cardioacceleratory effects of baroreceptor unloading even during normal heart rates in truly resting animals; preliminary experiments indicate that this is in fact the case (C. M. Wood, unpublished results).

The functional significance of bradycardia, especially hypoxic bradycardia, in fish has been much debated but remains unknown (cf. Shelton, 1970; Smith & Jones, 1978)

Much of this uncertainty derives from lack of knowledge of the influence of bradycardia on cardiac stroke volume and total $\dot{Q}$. The present results represent the first direct measurements of total $\dot{Q}$ during bradycardia in intact unanaesthetized fish. They clearly show that increases in stroke volume compensate for decreases in heart rate so that $\dot{Q}$ remains unchanged (spontaneous bradycardia) or actually increases slightly (hypoxic bradycardia). These observations agree with the conclusions of Holeton & Randall (1967*b*) from Fick experiments on trout and more recent Fick-based estimates on dogfish (Piiper, Baumgarten & Meyer, 1970; Butler & Taylor, 1975; Taylor, Short & Butler, 1977; Short, Butler & Taylor, 1977), all of which indicate compensating increases of stroke volume during hypoxic bradycardia. Thus the present findings support the interpretation that the significance of hypoxic bradycardia is that it makes the pattern of gill blood flow more efficient in gas transfer, rather than that it reduces total $\dot{Q}$ and thereby cardiac energy expenditure (cf. Shelton, 1970; Smith & Jones, 1978).

This inverse relationship between heart rate and cardiac stroke volume is probably largely attributable to Starling's Law, as discussed in detail by Taylor *et al.* (1977) and Short *et al.* (1977). A possible role for sympathetic stimulation of stroke volume either via direct innervation (Gannon & Burnstock, 1969; Gannon, 1971; Holmgren, 1977) or via circulatory catecholamines (Nakano & Tomlinson, 1967; Nilsson, Abrahamsson & Grove, 1976; Mazeaud, Mazeaud & Donaldson, 1977) cannot, however, be excluded. Positive sympathetic inotropism may be especially important during hypoxic bradycardia, where the rise in stroke volume more than compensated for the fall in heart rate and in fact persisted to some extent after heart rate had returned to normal (Fig. 3).

In conclusion, it appears that presso-disturbing stimuli such as sudden changes in R_s or blood volume can indirectly influence heart rate (and probably $\dot{Q}$) through effects on P_a that are detected by baroreceptors. The resulting changes in heart rate are mediated largely by alterations in vagal tone. This can occur even when vagal tone is under chemoreceptor drive, indicating a central interaction between the baroreceptor and chemoreceptor reflexes. The functional significance of the interaction is not clear. However, it is possible that the increased heart rate during depressor stimuli will raise $\dot{Q}$ to some extent. As R_s itself appear to be reflexly controlled via sympathetic vasomotor tone (Wood, 1974; Smith, 1978), the interactions and feedback between R_s, heart rate, $\dot{Q}$, and the setting of chemoreceptor and baroreceptor sensitivities in the teleost may well approach the complexity seen in mammals (Heymans & Neil, 1958; Öberg, 1976).

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REFERENCES

BERNE, R. M. & LEVY, M. N. (1972). *Cardiovascular Physiology*, 2nd ed. St Louis: C. V. Mosby.

BUTLER, P. J. & TAYLOR, E. W. (1975). The effect of progressive hypoxia on respiration in the dogfish (*Scyliorhinus canicula*) at different seasonal temperatures. *J. exp. Biol.* **63**, 117–130.

BURTON, A. C. (1972). *Physiology and Biophysics of the Circulation*, 2nd ed. Chicago: Yearbook Medical Publishers.

DAXBOECK, C. & HOLETON, G. F. (1978). Oxygen receptors in the rainbow trout, *Salmo gairdneri*. *Can. J. Zool.* **56**, 1254–1259.

GANNON, B. J. (1971). A study of the dual innervation of teleost heart by a field stimulation technique. *Comp. gen. Pharmac.* **2**, 175–183.

GANNON, B. J. & BURNSTOCK, G. (1969). Excitatory adrenergic innervation of the fish heart. *Comp. Biochem. Physiol.* **29**, 765–773.

HEISTAD, D. D., ABBOUD, F. M., MARK, A. L. & SCHMID, P. G. (1974). Interaction of baroceptor and chemoreceptor reflexes. Modulation of the chemoreceptor reflex by changes in baroreceptor activity. *J. clin. Invest.* **53**, 1226–1236.

HELGASON, S. & NILSSON, S. (1973). Drug effects on pre- and post-branchial blood pressure and heart rate in a free-swimming marine teleost, *Gadus morhua*. *Acta physiol. scand.* **88**, 533–540.

HEYMANS, C. & NEIL, C. (1958). *Reflexogenic Areas of the Cardiovascular System*. London: J. and A. Churchill.

HOLETON, G. F. & RANDALL, D. J. (1967*a*). Changes in blood pressure in the rainbow trout during hypoxia. *J. exp. Biol.* **46**, 297–305.

HOLETON, G. F. & RANDALL, D. J. (1967*b*). The effect of hypoxia upon the partial pressure of gases in the blood and water afferent and efferent to the gills of the rainbow trout. *J. exp. Biol.* **46**, 317–327.

HOLMGREN, S. (1977). Regulation of the heart of a teleost, *Gadus morhua*, by autonomic nerves and circulating catecholamines. *Acta physiol. scand.* **99**, 62–74.

MANCIA, G. (1975). Influence of carotid baroreceptors on vascular responses to carotid chemoreceptor stimulation in the dog. *Circulation Res.* **36**, 270–276.

MAZEAUD, M. M., MAZEAUD, F. & DONALDSON, E. M. (1977). Primary and secondary effects of stress in fish: some new data with a general review. *Trans. Am. Fish. Soc.* **106**, 201–212.

MOTT, J. C. 1951. Some factors affecting the blood circulation in the common eel (*Anguilla anguilla*). *J. Physiol., Lond.* **114**: 387–398.

NAKANO, T. & TOMLINSON, N. (1967). Catecholamine and carbohydrate metabolism in rainbow trout (*Salmo gairdneri*) in relation to physical disturbance. *J. Fish Res. Bd Can.* **24**, 1701–1715.

NILSSON, S., ABRAHAMSSON, T. & GROVE, D. J. (1976). Sympathetic nervous control of adrenaline release from the head kidney of the cod, *Gadus morhua*. *Comp. Biochem. Physiol.* **55C**, 123–127.

ÖBERG, B. (1976). Overall cardiovascular regulation. *Physiol. Rev.* **38**, 537–570.

PIIPER, J., BAUMGARTEN, D. & MEYER, M. (1970). Effects of hypoxia upon respiration and circulation in the dogfish *Scyliorhinus stellaris*. *Comp. Biochem. Physiol.* **36**, 513–520.

PRIEDE, I. G. (1974). The effect of swimming activity and section of the vagus nerves on heart rate in rainbow trout. *J. exp. Biol.* **60**, 305–319.

RANDALL, D. J. (1968). Functional morphology of the heart in fishes. *Am. Zool.* **8**, 179–189.

RANDALL, D. J. & SHELTON, G. (1963). The effects of changes in environmental gas concentrations on the breathing and heart rate of a teleost fish. *Comp. Biochem. Physiol.* **9**, 229–239.

RANDALL, D. J. & SMITH, J. C. (1967). The regulation of cardiac activity in fish in a hypoxic environment. *Physiol. Zoöl.* **40**, 104–113.

RANDALL, D. J. & STEVENS, E. D. (1967). The role of adrenergic receptors in cardiovascular changes associated with exercise in salmon, *Comp. Biochem. Physiol.* **21**, 415–424.

RISTORI, M. T. (1970). Réflexe de barosensibilité chez un poisson téléostéen (*Cyprinus carpio* L.). *C. r. séanc. Soc. Biol.* T **164**, 1512–1516.

RISTORI, M. T. & DESSAUX, G. (1970). Sur l'existence d'un gradient de sensibilité dans les recepteurs branchiaux de *Cyprinus carpio* L. *C. r. séanc. Soc. Biol.* T **164**, 1517–1519.

SHELTON, G. (1970). The regulation of breathing. In *Fish Physiology*, vol. IV (ed. W. S. Hoar and D. J. Randall). New York: Academic Press.

SHORT, S., BUTLER, P. J. & TAYLOR, E. W. (1977). The relative importance of nervous, humoral, and intrinsic mechanisms in the regulation of heart rate and stroke volume in the dogfish (*Scyliorhinus canicula*). *J. exp. Biol.* **70**, 77–92.

SMITH, D. G. (1978). Neural regulation of blood pressure in rainbow trout (*Salmo gairdneri*). *Can. J. Zool.* **56**, 1678–1683.

SMITH, F. M. & JONES, D. R. (1978). Localization of receptors causing hypoxic bradycardia in trout (*Salmo gairdneri*). *Can. J. Zool.* **56**, 1260–1265.

STEVENS, E. D., BENNION, G. R., RANDALL, D. J. & SHELTON, G. (1972). Factors affecting arterial

pressures and blood flow from the heart in intact, unrestrained lingcod, *Ophiodon elongatus*. *Comp. Biochem. Physiol.* **43**A, 681–695.

STEVENS, E. D. & RANDALL, D. J. (1967). Changes in blood pressure, heart rate, and breathing rate during moderate swimming activity in rainbow trout. *J. exp. Biol.* **46**, 307–315.

TAYLOR, E. W., SHORT, S. & BUTLER, P. J. (1977). The role of the cardiac vagus in the responses of the dogfish *Scyliorhinus canicula* to hypoxia. *J. exp. Biol.* **70**, 57–75.

WENNERGREN, G., LITTLE, R. & ÖBERG, B. (1976). Studies on the central integration of excitatory chemoreceptor influences and inhibitory baroreceptor influences and cardiac receptor influences. *Acta physiol. scand.* **96**, 1–18.

WOOD, C. M. (1974). Mayer waves in the circulation of a teleost fish. *J. exp. Zool.* **189**, 267–274.

WOOD, C. M. (1976). Pharmacological properties of the adrenergic receptors regulating systemic vascular resistance in the rainbow trout. *J. comp. Physiol.* B **107**, 211–228.

WOOD, C. M. (1977). Cholinergic mechanisms and the response to ATP in the systemic vasculature of the rainbow trout. *J. comp. Physiol.* B **122**, 325–345.

WOOD, C. M., PIEPRZAK, P. & TROTT, J. N. (1979). The influence of temperature and anaemia on the adrenergic and cholinergic mechanisms controlling heart rate in the rainbow trout. *Can. J. Zool.* **57**, 2440–2447.

WOOD, C. M. & SHELTON, G. (1975). Physical and adrenergic factors affecting systemic vascular resistance in the rainbow trout: a comparison with branchial vascular resistance. *J. exp. Biol.* **63**, 505–523.

WOOD, C. M. & SHELTON, G. (1980). Cardiovascular dynamics and adrenergic responses of the rainbow trout *in vivo*. *J. exp. Biol.* **87**, 247–270.