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Effects of Severe Hemorrhagic Hypotension on the Vasculature of the Chicken[1] (41412)

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Abstract. We have recently reported that hemorrhage to a mean arterial blood pressure (MABP) of 50 mm Hg in the chicken has no effect on total peripheral resistance or skeletal muscle vascular resistance (judged from changes in limb perfusion pressure (P_p) during constant blood flow). In the present study we report the effect of a more severe hemorrhagic hypotension (MABP = 25 mm Hg) on skeletal muscle vascular resistance in the constantly perfused hindlimb of the chicken following severance of the sciatic nerve trunk, bilateral cervical vagotomy, α-adrenergic blockade, or during artificial perfusion of the head with arterial blood. Concentrations of serotonin (SER), dopamine (DA), and norepinephrine (NE) in plasma were determined at different levels of hypotension. While hemorrhage to MABP = 50 mm Hg had no effect on P_p, a further hemorrhage to MABP = 25 mm Hg produced a sharp rise in P_p which was unaffected by severance of the sciatic nerve truck or bilateral cervical vagotomy. This vasoconstriction could be completely eliminated by intraarterial infusion of phentolamine or by pump perfusing the head during the hypotensive interval. Furthermore, concentrations of SER, DA, and NE were significantly elevated only when the rise in P_p was evident. We conclude that the vasoconstrictor response to severe hemorrhagic hypotension in the chicken is primarily mediated by an increase in circulating catecholamines due to cerebral ischemia, rather than a baroreflex.

It has been demonstrated in most mammalian species that acute blood loss is immediately followed by reductions in cardiac output and arterial blood pressure. The most prominent and immediate compensatory response is reflex activation of the autonomic nervous system (1–3). It is this system that elevates vascular resistance and produces positive inotropic and chronotropic cardiac effects. The ability of mammals and flying or diving avian species to withstand a large acute blood loss with a minimal reduction in mean arterial blood pressure (MABP) is largely attributed to the efficacy of the sympathicoadrenal system to initiate an increase in precapillary resistance (1–3). A small hemorrhage in the domestic chicken (*Gallus domesticus*), on the other hand, is associated with a large fall in MABP (4, 5). This is likely due to a lack of sympathetic activation in this species, since a moderate hemorrhage (MABP = 50 mm Hg) is not associated with a rise in total peripheral resistance or skeletal muscle vascular resistance (5). In the present study, we examine the effect of a more severe hemorrhage (MABP = 25 mm Hg) on skeletal muscle vascular resistance in the isolated constantly perfused chicken leg.

Materials and Methods. Four experimental series were performed using 26 adult chickens of both sexes. The chickens were anesthetized with sodium pentobarbital (25 mg/kg iv), tracheotomized, artificially ventilated, and heparinized systemically (390 IU/kg). Arterial blood pressure was monitored from a cannula inserted into a carotid or brachial artery connected to a Statham transducer (PA-23AC) attached to a Grass 7A polygraph. Body temperature was maintained at 41° by a heating pad under the animal. The right ischiadic artery was cannulated for hemorrhaging and reinfusing the animals. In some animals, the sciatic nerve of the perfused limb or the vagi were isolated and looped with suture for subsequent

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sectioning. In the first three series, the blood supply to the leg was isolated, and the limb was pump perfused with arterial blood at a constant rate via a polyethylene cannula inserted midthigh in a cranial direction into the left ischiadic artery. Blood was then shunted to the perfusion pump and returned to the same artery through a cannula inserted caudally. The perfusion pressure (P_p) was measured in the pump outflow line. The P_p varied directly with vascular resistance since blood flow was constant throughout the duration of the experiment. The perfusion pump was turned off and a $P_p < 25$ mm Hg indicated adequate vascular isolation. For further description of this technique see reference (5).

The six female chickens of series 1 (1.84 ± 0.07 kg) were hemorrhaged to a MABP of 50 mm Hg and the P_p was recorded. The MABP was held at 50 mm Hg by continuous small bleedings. When all values had stabilized the animal was further hemorrhaged to a MABP = 25 mm Hg and was maintained at that level of hypotension by subsequent small bleedings and the P_p was again recorded. When the P_p stabilized the sciatic nerve trunk was severed and the P_p was monitored. Next, the α-adrenergic antagonist phentolamine was infused into the perfusion line proximal to the perfusion pump (50 μg/min). The P_p was monitored until it stabilized at which time the phentolamine infusion was discontinued and the shed blood was returned to the animal.

In series 2, seven hens (1.82 ± 0.07 kg) were hemorrhaged to a MABP of 50 mm Hg and then 25 mm Hg while P_p was monitored as in the previous series. The shed blood was returned and a bilateral cervical vagotomy was performed and the animals were again hemorrhaged to a MABP of 25 mm Hg while the P_p was recorded.

Series 3 used eight adult male chickens (2.33 ± 0.06 kg) with a leg perfusion preparation similar to the first two series except that the head of the animal was also pump perfused. This was done by pumping (Sigma motor pump) blood from an ischiadic artery through a bifurcated cannula inserted in a cranial direction into both carotid arteries. The carotid perfusion pressure was monitored in the pump outflow line. MABP was monitored from a brachial artery and the animal was hemorrhaged via a cannula inserted caudally into a carotid artery. The shed blood was reinfused via a cannula inserted caudally into an ischiadic artery. The carotid perfusion pump was turned off and the animal was hemorrhaged to a MABP which produced a large increase in P_p. The shed blood was then reinfused, the carotid perfusion pump was turned back on, and the animal was allowed to stabilize. The animal was again hemorrhaged to the same MABP as before but the carotid perfusion pump continued to perfuse the head. The carotid perfusion pressure was monitored during both hemorrhages.

In the fourth experimental series, five male chickens (1.96 ± 0.14 kg) were held at a MABP of 50 mm Hg for 30 min and then at a MABP of 25 mm Hg for 30 min by continuous small bleedings. Arterial blood samples (1 ml) were drawn prior to hemorrhage, at the 5th, 15th, and 30th min of each level of hypotension, and at 10 min following reinfusion of the shed blood. The blood was centrifuged and the plasma was frozen for subsequent analysis for serotonin, dopamine, and norepinephrine concentrations by a slight modification of the method of Jacobowitz and Richardson (6).

All values are given as mean ± SEM. Statistical analysis was via Student's t test. The changes in plasma hormone concentration over time were compared with the initial value via a one way ANOVA with a Dunnett test. A $P < 0.05$ was considered significant.

Results. Table I summarizes the results of the first three experimental series. In all animals, a hemorrhage to a MABP of 50 mm Hg did not produce a change in P_p, whereas a hemorrhage to 25 mm Hg produced a significant increase in P_p. The P_p immediately began to increase as the MABP fell near 25 mm Hg and it remained elevated until the shed blood was reinfused. During reinfusion of the shed blood, the P_p would decrease to the control value following the return of enough blood to raise the MABP to approximately 50 mm Hg.

In the first series, severance of the sciatic

TABLE I. THE EFFECT OF HEMORRHAGE ON MEAN ARTERIAL BLOOD PRESSURE (MABP), HINDLIMB PERFUSION PRESSURE (HPp), AND CAROTID PERFUSION PRESSURE (CPp) IN 21 ADULT CHICKENS FOLLOWING VARIOUS EXPERIMENTAL CONDITIONS

Condition	MABP (mm Hg)	HPp (mm Hg)	CPp (mm Hg)
Series 1 (n = 6)			
Control	124 ± 6	97 ± 5	—[a]
Hemorrhage	50 ± 2	97 ± 5	—
Hemorrhage	25 ± 2	174 ± 10*	—
Hemorrhage + sciatic[b]	25 ± 2	172 ± 11*	—
Hemorrhage + phentol.[c]	25 ± 2	92 ± 10	—
Series 2 (n = 7)			
Control	109 ± 6	77 ± 8	—
Hemorrhage	50 ± 2	77 ± 8	—
Hemorrhage	25 ± 2	189 ± 9*	—
Control	111 ± 4	79 ± 9	—
Hemorrhage + vagot.[d]	25 ± 2	184 ± 10*	—
Series 3 (n = 8)			
Control	109 ± 6	144 ± 7	141 ± 7
Hemorrhage + CPP[e] off	37 ± 2	335 ± 20*	19 ± 2
Control	121 ± 9	153 ± 12	157 ± 8
Hemorrhage + CPP on	35 ± 2	174 ± 15	96 ± 12

Note. All values expressed as mean ± SE.
[a] CPp not measured in series 1 and 2.
[b] Severance of the sciatic nerve trunk during hemorrhage.
[c] Intraarterial infusion of phentolamine (50 μg/min) during hemorrhage.
[d] Bilateral cervical vagotomy prior to hemorrhage.
[e] CPP designates carotid perfusion pump.
* Significant change in HPp from the preceding control ($P < 0.05$).

nerve during the elevated P_p would generally produce only a transient (30 sec) 25–30 mm Hg fall in P_p, other than this the P_p was unaffected (Table I). In contrast, an intraarterial infusion of phentolamine would promptly return the P_p to the prehemorrhage level, where it was maintained as long as the α-blocker was infused.

In the second series, the P_p again was unaffected by a hemorrhage to 50 mm Hg and increased significantly as the MABP was reduced to 25 mm Hg (Table I). MABP and P_p returned to control values following reinfusion of the shed blood. The rise in P_p in response to hemorrhage was unaltered by bilateral vagotomy.

In the third series, a hemorrhage to a MABP of 25 mm Hg was not required to produce a rise in limb P_p (Table I). Indeed in some birds there was intense vasoconstriction in the limb at a MABP of 45 mm Hg. However, at this time carotid pressure was <25 mm Hg. When cerebral blood flow was artificially maintained during hemorrhage, no rise in skeletal muscle P_p, i.e., vascular resistance, occurred (Table I). Figure 1 is a representative tracing showing the effect of hemorrhage on limb P_p with and without head perfusion. It is evident that P_p increased markedly, i.e., 150 mm Hg, when carotid pressure fell below 25 mm Hg even though MABP was 40 mm Hg. However, when head flow was maintained, the limb P_p rose only 20 mm Hg during the hypotensive period.

The results of the fourth series are shown in Fig. 2. A hemorrhage to a MABP of 50 mm Hg did not effect the concentration of serotonin, dopamine, or norepinephrine in

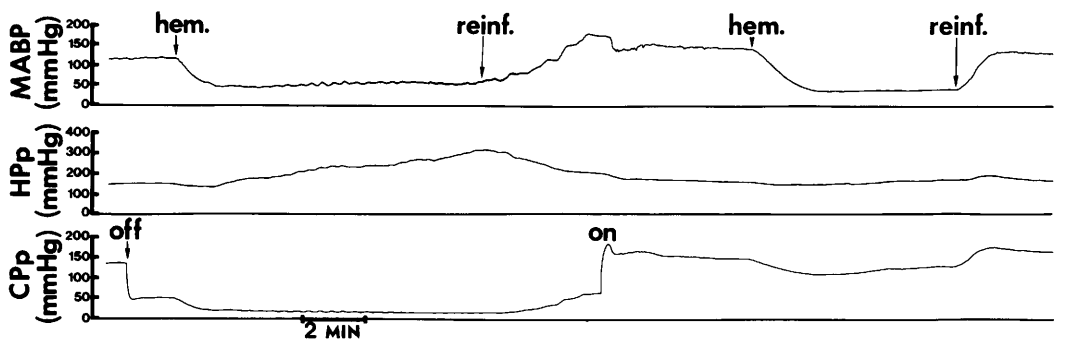

FIG. 1. Continuous tracing of mean arterial blood pressure (MABP), hindlimb perfusion pressure (HPp), and carotid perfusion pressure (CPp) in the chicken (off, carotid perfusion pump off; hem, hemorrhage; reinf, reinfusion of shed blood; on, carotid perfusion pump on).

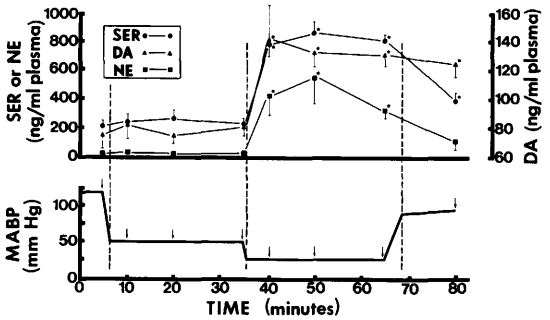

FIG. 2. The effect of various levels of hemorrhagic hypotension on the concentrations of serotonin (SER), dopamine (DA), and norepinephrine (NE) in plasma of chickens (n = 5). Values expressed as mean ± SEM. Asterisk indicates significant change from control ($P < 0.05$).

plasma. However, the concentrations of all three hormones increased significantly when the MABP was lowered to 25 mm Hg. The initial MABP in this series was 127 ± 15 mm Hg.

Discussion. The finding that small reductions in blood volume are associated with marked hypotension has previously been reported in the chicken (4, 5). We recently reported that although α-adrenergic receptors are present in the vasculature, they are seemingly not activated to a measurable extent by the stress of a hemorrhage to a MABP of 50 mm Hg (5). However, a mild cerebral ischemic response (increase in MABP) has been reported in chickens after bilateral occlusion of the carotid and vertebral arteries (7) and intense vasoconstriction of the skeletal muscle vasculature has recently been demonstrated in chickens during asphyxia (8). In the present study, severe hemorrhage produced a similar vasoconstriction, again mediated through activation of the α-receptors (blocked by the α-antagonist phentolamine). The fact that the vasoconstriction was not eliminated, or even attenuated, by bilateral vagotomy suggests that the response to severe hypotension is not mediated via the baroreflex. The chicken, unlike the mammal, does not have a functional carotid sinus baroreceptor (7).

The vasoconstrictor response is apparently due largely to an adrenal catecholamine release inasmuch as severance of the sciatic nerve, which most likely contains some sympathetic efferents to the skeletal muscle vasculature under study, only transiently attenuated the vasoconstriction. Other researchers have also suggested that, in the chicken, medullary hormones play an important role in regulating cardiac performance and blood pressure (9, 10). The control concentrations of norepinephrine and serotonin are similar to those previously reported in the chicken (10–12). The dramatic increase in the concentration of catecholamines, which occurs at the time of the rise in P_p, also supports the hypothesis that the vasoconstriction is predominantly the result of humoral pressor agents rather than activation of sympathetic efferents. During the carotid perfusion studies, the vasoconstrictor response in the limb was independent of MABP, suggesting it was not initiated via aortic baroreceptors. We conclude that the vasoconstriction is not part of an autonomic baroreflex, but rather, it is due to the release of adrenal medullary hormones in response to cerebral ischemia.

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