Bulbar and Suprabulbar Control of the Cardiovascular Autonomic Effects during Arterial Hypoxia in the Rabbit

By John B. Uther, M.B., Stephen N. Hunyor, M.B., John Shaw, M.B., and Paul I. Korner, M.D.

ABSTRACT

The autonomic effects on heart rate, portal, renal, cutaneous and muscle blood flows were studied during approximately constant severe arterial hypoxia in unanesthetized sham-operated, thalamic and pontine rabbits, 3 hours after operations conducted under halothane anesthesia. Neural and adrenal effects were estimated by comparing the responses of different groups of animals: (a) with all neural effectors intact, (b) after adrenalectomy, (c) after selective and (d) after complete pharmacological block of neural effector mechanisms. In pontine animals hypoxia greatly increased cardiac sympathetic activity and seemed to evoke a greater release of adrenal catecholamine (epinephrine) than in other preparations, but caused significant neural peripheral sympathetic constrictor effects in only the portal bed. In thalamic animals, hypoxia caused reflex bradycardia, striking neural constrictor effects in the portal, renal and muscle beds, and cutaneous dilatation. There was little evidence of an increase in epinephrine secretion. It is suggested that there is an inhibitory pathway between diencephalon and bulb that limits the release of epinephrine. Sham-operated animals exposed to hypoxia secreted epinephrine which played an important role in the total reflex response; this group had similar cardioinhibitory and cutaneous effects to thalamic animals, but a more rapid onset of portal and renal constriction and a marked increase in muscle blood flow. The sympathetic responses of the different preparations resulted in different rates of metabolic H+ ion production.

ADDITIONAL KEY WORDS forebrain adrenal catecholamines diencephalic and pontomedullary circulatory centers arterial chemoreceptors portal, renal, muscle, skin blood flow

Although cortical and diencephalic structures (1, 2) are known to influence circulatory changes that occur in muscular exercise (3), emotion (4), and thermoregulation (5), their role in reflexes from cardiovascular receptors has only recently been recognized (6). Previous results in unanesthetized rabbits have shown that during the reflex activity evoked by arterial hypoxia, suprabulbar structures contribute to the bulbar reflex autonomic effects on heart rate and total peripheral resistance (7). We have examined here the contribution of these suprabulbar mechanisms to the activity of the main autonomic effectors, by studying changes in heart rate, and in portal, renal, muscle and cutaneous blood flows in sham-operated, thalamic and pontine preparations during severe arterial hypoxia. We have assessed the relative importance of neural and adrenal effects in these several preparations by comparing the hypoxic responses of animals with all effectors intact.
with those of adrenalectomized animals and those of animals subjected either to selective or to complete autonomic block.

**Methods**

The experiments were performed on male white rabbits of mean body weight 2.6 kg (range 2.3 to 3.6 kg) of the same strain as used previously (7).

**Operations.**—To measure peripheral blood flow, local thermodilution catheters were inserted under halothane anesthesia into the left renal, portal and common iliac veins 7 to 10 days before an experiment (8, 9). In some animals bilateral adrenalectomy was performed at the same time (10) and in these a renal vein catheter was not inserted, since the adrenalectomy itself destroyed some of the renal nerves. All adrenalectomized animals were subsequently maintained on fixed steroid replacement therapy with cortisone acetate (1.0 mg/day im) and deoxycorticosterone acetate (1.5 mg/day im); these doses are adequate to maintain normal food and water consumption, serum Na and K levels, blood urea and postoperative hematocrits close to the preoperative values (10). Some of the adrenalectomized animals were further treated with guanethidine and atropine to inactivate the remaining autonomic effectors. Such autonomically "de-efferented" adrenalectomized animals nevertheless retained an essentially normal respiratory response to arterial hypoxia (7).

On the day of the experiment the central ear artery and right atrium were cannulated, and a tracheotomy tube was inserted under local lidocaine anesthesia.

The neurosurgical procedures were then performed under open circuit halothane anesthesia (7). After a frontoparietal craniotomy, appropriate regions of the brain were removed with minimal blood loss (3 to 8 ml) by diathermy suction. The preparations comprised (1) sham-operated animals in which bilateral craniotomy was performed and 5 ml of blood was slowly removed through the arterial catheter during anesthesia (extended to 1 hour, the approximate duration of the other operations); (2) thalamic animals, in which the cerebral hemispheres including basal ganglia were removed, but the thalamus and hypothalamus were preserved intact (Fig. 1); (3) pontine animals, in which the brain was removed rostral to a plane from the lower border of the inferior colliculus to the anterior ventral border of the pons just behind the point of emergence of the oculomotor nerves (Fig. 1). The animals recovered in a sound-proof box and were given 15 ml of 5.5% dextrose iv every hour. The sham-operated and thalamic animals regained their normal posture and movements within 30 to 40 minutes after operation (7). In the pontine animals body temperature was maintained postoperatively by an electric blanket. The animals lay on their sides until half an hour before the experiment, when they were placed into a special holder and studied in the posture of a normally sitting rabbit.

**Drugs.**—Atropine was administered 10 to 15 minutes before an experiment in an initial dose of 3 mg atropine sulfate iv, followed by 0.2 mg/min iv by continuous infusion. This dose is adequate to block heart rate changes after electrical stimulation of the vagus in the anesthetized rabbit (11). Propranolol was given in a dose of 0.4 mg/kg/10 min iv, a dose adequate to block the tachycardia and fall in blood pressure normally produced by the rapid injection of 4 μg isoproterenol sulfate iv. To produce autonomic de-efferentation, guanethidine sulfate was administered to adrenalectomized rabbits 4 days after operation in a dose of 12.5 mg/kg/day iv for 4 to 7 days. This regime blocks sympathetic nerve transmission and depletes tissue catecholamine stores (12).

**Cardiorespiratory Measurements.**—Portal, renal and iliac vein blood flows were measured by local thermodilution, after a rapid injection of 0.45 ml of 5.5% dextrose in distilled water at room temperature 11 to 12 mm upstream from the

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**Figure 1**

Diagram illustrating extent of brain removal in thalamic and pontine preparations.
CENTRAL CONTROL OF REFLEX AUTONOMIC RESPONSE

100 mm Hg.

Portal

°C

0

mm Hg.

Portal

°C

0

10 Sec.

Portal

°C

0

100 mm Hg.

Portal

°C

0

FIGURE 2

Top: Records of local thermodilution or thermal conductivity curves obtained in the resting sham-operated rabbit from A, portal vein; B, common iliac vein; right (R) and left (L) ankle skin and ear (E) skin. Records from top: cutaneous heat flow baseline, arterial pressure, right atrial pressure, time-temperature curve, integrated time-temperature curve.

Bottom: C, Portal thermodilution curve and D, iliac thermodilution and heat flow curves obtained 10 minutes after onset of hypoxia, showing reduction in portal flow and increase in iliac vein flow, and increase in limb skin and ear heat flow.

thermistor (8, 9) (Fig. 2). Blood flows in ankle skin and ear skin were estimated by a thermal conductivity method, calibrated in separate perfusion experiments relating heat flow to cutaneous blood flow (13). Muscle blood flow was estimated as the difference between simultaneously recorded common iliac vein blood flow and total hindlimb skin blood flow; bone blood flow and venous drainage from pelvic contents, which mostly drain into the tail vein (9), were neglected. The accuracy of the thermodilution method for measuring venous blood flow and the assumptions and limitations necessary to estimate muscle blood flow from measurements of iliac vein flow and cutaneous heat flow, and the measurement of changes within animals in the various regional blood flows have been discussed previously (8, 12).

Mean ear artery and right atrial pressures were recorded by Statham P 23 AC strain gauges. Heart rates were obtained from the arterial pressure pulse. In the peripheral beds vascular "resistance" was calculated as the ratio of ear artery pressure to peripheral blood flow, neglecting local venous pressure, since pressure recording through the slot of the injection lumen of the local thermodilution catheters was unsatisfactory.

The animals breathed room air through a low-resistance flap valve during resting and recovery periods; during the test period a freshly prepared low O2-N2 mixture (7.5 to 8% O2) was administered from a light latex balloon (7, 12). Respiration rate was measured by counting respiratory movements, and minute ventilation (1/min ATPS) was measured by collecting the expired air. Arterial Po2, Pco2 and pH were measured by a blood gas analyzer and pH meter (Instrumentation Laboratory Inc. Model 113).

Experimental Procedures.—Except in autonomically de-afferented animals not subjected to the sham operation, the experiment began 3 hours from the end of anesthesia, with the animal inside the sound-proof box. The experiments consisted of a resting period of 12 minutes in which the rabbit breathed room air (5 sets of circulatory and 2 respiratory measurements), followed by a period of 45 minutes during which it breathed 7.5 to 8% O2 in N2 (16 circulatory and 9 respiratory sets of measurements), followed by a 12-minute recovery period in which it breathed room air (4 sets of
circulatory and 2 respiratory measurements) (Fig. 3). One set of measurements of arterial P<sub>ao</sub><sub>2</sub>, P<sub>co</sub><sub>2</sub> and pH was obtained during the resting period while the rabbit breathed room air, and two others 12 and 32 minutes after the onset of hypoxia.

Presentation of Results and Statistical Meth...

ods.—The timing of the measurements was identical in all experiments. In each group the 25 sets of observations from the various animals were averaged at each time interval (Fig. 3A), and the standard error of the mean at a single time interval was calculated by analysis of variance after subtraction of between-animals and between-times sums of squares from the total sum of squares, as (error mean square/n) where n is the number of animals in the group. The means of the 25 sets of measurements were then grouped into 11 larger time intervals, as shown in Figure 3B. The standard error of the mean of a larger time interval was SE/(N), where N is the number of measurements per animal involved in each larger grouping. The standard error of the resting value was based on 5 measurements per animal, and all other intervals were based on 2 measurements per animal except for the first interval from the start of hypoxia (1 measurement) and the penultimate one before the end of hypoxia (3 measurements) (Fig. 3B). For a comparison of the difference in response at any given time interval between two different groups of n<sub>1</sub> and n<sub>2</sub> animals, with standard errors se<sub>1</sub> and se<sub>2</sub> for that time interval, the standard error of the difference is (se<sub>1</sub><sup>2</sup> + se<sub>2</sub><sup>2</sup>)<sup>1/2</sup> with (n<sub>1</sub> + n<sub>2</sub> - 2) degrees of freedom. Significance of differences between time intervals was determined by variance ratio or by t-test (14).

The absolute P<sub>ao</sub><sub>2</sub> during hypoxia, rather than the change from resting, was regarded as the better measure of chemoreceptor stimulus, since chemoreceptor activity during inhalation of air varies relatively little in the range of P<sub>ao</sub><sub>2</sub> 118 to 90 mm Hg which occurs between different animals in the various groups. Therefore in these experiments attainment of a P<sub>ao</sub><sub>2</sub> of 30 to 35 mm Hg during hypoxia was regarded as the primary objective, and the standard errors after each P<sub>ao</sub><sub>2</sub> indicate the range of variation at each time between different animals (Table 3). On the other hand changes in P<sub>co</sub><sub>2</sub> and pH from resting can be considered as “secondary” effects of the low P<sub>ao</sub><sub>2</sub> and the standard errors in Table 3 after each variable were calculated to allow estimation of the significance of differences within animals in these variables by analysis of variance from the error mean square after removal of between-times and between-animals from the total sums of squares.

Assumptions and Limitations.—The present analysis is based on comparisons of the responses of groups of 3 to 10 animals of the different preparations. We have found in rabbits of the same strain, sex, approximate age and weight that the responses to hypoxia are reproducible within statistically defined error limits as in Figure 3C and D. In addition, the responses of heart rate...
and arterial and right atrial pressures to hypoxia in sham-operated, thalamic and pontine preparations in the present study were closely similar to those of corresponding preparations studied previously (7).

The use of acute preparations subjected to ablation of different parts of the brain permits an assessment of the function of the part of the brain remaining after ablation. Whether the residual centers function in the same way in the intact animal will depend on the degree of nonspecific damage produced by the preceding surgery and on abnormalities in the profile of stimulation of the different cardiovascular receptors. Previous data have indicated that nonspecific effects from surgery on resting respiration, heart rate, arterial pressure and cardiac output were small (7), and in the present study differences in the resting blood flow measurements between sham-operated, thalamic and pontine animals were also slight. The resting values of sham-operated animals were closely similar to results in normal animals not subjected to craniotomy and bleeding (15) except for the muscle blood flow, which was lower, presumably because of the longer period of relative inactivity in the sham-operated group, and we have assumed in our analysis that any nonspecific effect of neurosurgery or sham operation on vascular or autonomic responsiveness will affect all preparations equally.

In studying the different preparations following adrenalectomy, the animals were given a normal dose of steroid replacement on the day of the experiment. These animals withstood the neurosurgical procedures and subsequently hypoxia without ill effect, and their resting heart rate, blood pressures and muscle and cutaneous blood flows were similar to the corresponding preparations with adrenals intact (P < 0.01), and this probably was a nonspecific effect of surgical procedures. Thalamic animals with intact adrenals and adrenalectomized thalamic animals showed much the same portal vasoconstriction during hypoxia, which suggests that the intrinsic sensitivity of these beds was not greatly altered in the adrenalectomized group on the present regime of fixed steroid replacement, and that the nonspecific effect of the operation on resting portal blood flow was not itself an important factor modifying vascular responsiveness during hypoxia.

Autonomically de-efferented animals were used to assess the local peripheral effects of hypoxia. The completeness of inactivation of the autonomic effectors, the retention of a normal respiratory response and the assumption that within limits these local effects are approximately similar in animals with autonomic effectors intact have been discussed previously (12), as well as the reasons for not subjecting the animals to preliminary operation and anesthesia (7, 11).

In using atropine to estimate cardiac autonomic effects, we have assumed that it mainly blocks vagal effects and have neglected its effects on cardiac sympathetic ganglionic transmission (16) and any possible effects on arterial chemoreceptor discharge, in view of the relatively slight effect of atropine in modifying the heart rate response of the pontine animal (Fig. 5).

Results

**RESTING RABBITS BREATHING AIR**

The circulatory data obtained under these conditions are summarized in Table 1. In the various preparations with intact adrenals (termed normal preparations) heart rate and portal and limb skin blood flows were significantly lower in normal pontine animals than in the normal sham-operated and thalamic groups. In adrenalectomized preparations there was an increase in portal blood flow, probably non-specific (see Assumptions and Limitations), but the other variables were closely similar to corresponding values in normal animals. In sham-operated and pontine groups, resting cardiac sympathetic and vagal tone were demonstrated by measuring the changes in heart rate before and after propranolol or atropine. The effects of these drugs were not studied in thalamic animals. The resting ventilation volumes (Table 2) and the blood gas tensions (Table 3) of the normal sham-operated and pontine animals were very similar. As noted previously (7), the thalamic animals showed a higher resting ventilation, and this of course was reflected in a raised arterial Po2 and a lowered arterial PCO2.

**EFFECTS OF Pao2 30 TO 35 mm Hg**

*Respiration and Arterial Blood.*—All groups tolerated hypoxia well and exhibited only slight disturbances of behavior with some restlessness during the first 1 to 2 minutes of hypoxia (7). As observed previously (7), the respiratory responses of normal sham-operated and pontine animals were similar, but the increase in ventilation in thalamic animals was significantly smaller (Table 2). These re-
### TABLE 1

**Mean Values of the Cardiovascular Variables in the Resting Preparations**

<table>
<thead>
<tr>
<th>Group</th>
<th>Heart rate (beats/min)</th>
<th>Arterial pressures (mm Hg)</th>
<th>R. atrial pressures (mm Hg)</th>
<th>Portal blood flow (ml/min)</th>
<th>Renal blood flow (ml/min)</th>
<th>Muscle blood flow (ml/min/blind limb)</th>
<th>Limb skin blood flow (ml/min/100 g)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Sham-operated</strong></td>
<td></td>
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</tr>
<tr>
<td>Normal*</td>
<td>302 ± 0.5</td>
<td>93 ± 2.1</td>
<td>-0.8 ± 0.27</td>
<td>105 ± 15.5</td>
<td>57 ± 6.8</td>
<td>14 ± 3.2</td>
<td>12 ± 0.6</td>
</tr>
<tr>
<td>(10)</td>
<td>(10)</td>
<td>(10)</td>
<td>(9)</td>
<td>(9)</td>
<td>(6)</td>
<td>(6)</td>
<td>(9)</td>
</tr>
<tr>
<td>Adrenalectomy</td>
<td>291 ± 0.7</td>
<td>87 ± 3.7</td>
<td>-0.5 ± 0.34</td>
<td>134 ± 13.9†</td>
<td>21 ± 4.3</td>
<td>10 ± 0.5</td>
<td>18 ± 3.3</td>
</tr>
<tr>
<td>(9)</td>
<td>(9)</td>
<td>(9)</td>
<td>(8)</td>
<td>(7)</td>
<td>(7)</td>
<td>(7)</td>
<td></td>
</tr>
<tr>
<td>Propranolol</td>
<td>228 ± 10.6‡</td>
<td>97 ± 4.7</td>
<td>-0.1 ± 0.43</td>
<td>100 ± 11.1</td>
<td>19 ± 3.5</td>
<td>11 ± 0.6</td>
<td>14 ± 0.3</td>
</tr>
<tr>
<td>(5)</td>
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<td>(3)</td>
<td>(3)</td>
<td>(3)</td>
<td>(5)</td>
</tr>
<tr>
<td>Atropine</td>
<td>335 ± 12.9§</td>
<td>87 ± 2.2</td>
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<td>(7)</td>
<td>(7)</td>
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<tr>
<td><strong>Thalamic</strong></td>
<td></td>
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</tr>
<tr>
<td>Normal</td>
<td>295 ± 10.5</td>
<td>91 ± 4.6</td>
<td>-0.9 ± 0.74</td>
<td>95 ± 13.8</td>
<td>70 ± 10.4</td>
<td>24 ± 6.0</td>
<td>10 ± 1.2</td>
</tr>
<tr>
<td>Adrenalectomy</td>
<td>323 ± 17.0</td>
<td>85 ± 3.8</td>
<td>-1.8 ± 0.71</td>
<td>129 ± 21.2†</td>
<td>16 ± 4.5</td>
<td>12 ± 0.3</td>
<td>16 ± 1.8</td>
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<td>(4)</td>
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<tr>
<td><strong>Pontine</strong></td>
<td></td>
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</tr>
<tr>
<td>Normal</td>
<td>246 ± 10.9</td>
<td>89 ± 3.9</td>
<td>+0.1 ± 0.46</td>
<td>62 ± 7.3‡</td>
<td>52 ± 8.8</td>
<td>26 ± 8.8</td>
<td>8 ± 0.43</td>
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<td>(6)</td>
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<td>(6)</td>
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<tr>
<td>Adrenalectomy</td>
<td>248 ± 5.4</td>
<td>93 ± 2.0</td>
<td>+0.1 ± 0.30</td>
<td>104 ± 16.0‡</td>
<td>25 ± 6.3</td>
<td>7 ± 0.4</td>
<td>13 ± 0.1</td>
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<td>Propranolol</td>
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<td>79 ± 4.2</td>
<td>-0.3 ± 0.27</td>
<td>98 ± 12.4</td>
<td>23 ± 6.0</td>
<td>9 ± 1.2</td>
<td>13 ± 0.0</td>
</tr>
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<td>(3)</td>
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<td>(3)</td>
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<tr>
<td>Atropine</td>
<td>262 ± 5.6§</td>
<td>93 ± 1.7</td>
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</tr>
<tr>
<td>De-efferented</td>
<td>230 ± 3.6</td>
<td>82 ± 4.6</td>
<td>+1.0 ± 0.20</td>
<td>117 ± 13.5</td>
<td>52 ± 9.0</td>
<td>24 ± 5.0</td>
<td>10.5 ± 0.5</td>
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<td>(6)</td>
<td>(6)</td>
<td>(6)</td>
<td>(3)</td>
<td>(3)</td>
<td>(6)</td>
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</table>

*Values are means ± s.e; figure in parentheses is number of rabbits.
†Portal blood flow after adrenalectomy significantly higher than in preparations with adrenals intact, determined in separate analysis of variance (P = 0.01).
‡Mean heart rate within animals before propranolol, 295 in sham-operated and 250 in pontine animals; P < 0.01 for significance of difference due to the drug in both groups.
§Normal pontine portal blood flow significantly below normal sham-operated or thalamic animals, or propranolol-treated pontine animals (P < 0.02).
¶Mean heart rate within animals before atropine, 290 in sham-operated and 229 in pontine animals; P < 0.05 for significance of difference due to the drug.
TABLE 2
Mean Respiratory Values before, during and after Hypoxia

<table>
<thead>
<tr>
<th>Groups</th>
<th>No. of rabbits</th>
<th>Variable</th>
<th>Resting*</th>
<th>Hypoxia*</th>
<th>Recovery*</th>
<th>SE†</th>
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<tr>
<td>Normal†</td>
<td>10</td>
<td>VE</td>
<td>1.16</td>
<td>2.33</td>
<td>1.62</td>
<td>±0.087</td>
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<tr>
<td></td>
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<td>f</td>
<td>65</td>
<td>92</td>
<td>71</td>
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<tr>
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<td>9</td>
<td>VE</td>
<td>0.92</td>
<td>1.95</td>
<td>1.18</td>
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<td></td>
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<td>108</td>
<td>74</td>
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<td>1.69</td>
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<td></td>
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<td>74</td>
<td>89</td>
<td>73</td>
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<td>112</td>
<td>82</td>
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<td>±2.3</td>
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<td>88</td>
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</tbody>
</table>

*In each animal the values at the different periods were obtained as follows:—(1) Resting, mean of two read-
ing, at beginning and end; (2) Hypoxia, mean of nine readings from ninth to forty-fifth minute, when
there is an approximate steady-state at this Pao2 (7); (3) Recovery, mean of two readings 9 and 12 minutes
from end of hypoxia.

†Values are means for ventilation (VE) and breaths/minute (f); se values determined by analysis of variance
and based on within-animal comparisons to estimate the significance of any changes.

§Change from resting in VE during hypoxia significantly smaller than in normal sham-operated or pontine ani-
mals (P < 0.01).

$Resting VE higher than in normal sham-operated animal; se diff. 0.21; P < 0.05.

responses were little altered after adrenalectomy. In sham-operated and pontine animals
 treated with propranolol, the rise in ventilation caused by hypoxia was less marked than
 without the drug (P < 0.001).

Pao2 and Paco2 during hypoxia were closely similar in the different groups (Table 3). The
changes in arterial blood pH in normal sham-operated and thalamic animals were small; pH
decreased by 0.06 ± 0.031 (se) units (P = 0.1) and 0.13 ± 0.059 units (0.1 > P > 0.05)
after 32 minutes of hypoxia. In the normal pontine group hypoxia caused a significant fall
of 0.24 ± 0.055 units (P < 0.02). In adrenalectomized and propranolol-treated sham-op-
erated and pontine preparations, the arterial pH rose significantly during hypoxia, suggest-
ing that adrenal catecholamines had contributed through their beta-adrenergic effects to
the production in H+ ions during hypoxia masking the expected respiratory alkalosis. In
the thalamic preparation there was no difference between the arterial pH response of normal
and adrenalectomized animals; in both groups there was masking of the expected respiratory alkalosis, but it would seem from the results that the adrenals contributed little if anything to the production of H+ ions in this
preparation.

Heart Rate and Blood Pressure.—In normal sham-operated and thalamic animals, the
heart rate fell to a minimum of about 60% of the resting value 1 minute after the start of hypoxia, remained significantly reduced
### TABLE 3

Mean Values of Arterial Oxygen Pressure, Carbon Dioxide Pressure, and pH during Resting Period and Breathing Room Air and during Hypoxia

<table>
<thead>
<tr>
<th>Group</th>
<th>No. of rabbits</th>
<th>Arterial O₂ pressure* (mm Hg)</th>
<th>Arterial CO₂ pressure (mm Hg)</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Air 12 min 32 min Hypoxia</td>
<td>Air 12 min 32 min SE†† Hypoxia</td>
<td>Air 12 min 32 min SE†† Hypoxia</td>
</tr>
<tr>
<td>Sham-operated</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Normal</td>
<td>10</td>
<td>105 ± 1.0</td>
<td>32 ± 0.4</td>
<td>25 ± 16</td>
</tr>
<tr>
<td>Adrenalectomy</td>
<td>9</td>
<td>95 ± 1.3</td>
<td>30 ± 0.44</td>
<td>29 ± 17</td>
</tr>
<tr>
<td>Propranolol</td>
<td>5</td>
<td>95 ± 3.6</td>
<td>31 ± 1.16</td>
<td>32 ± 17</td>
</tr>
<tr>
<td>Atropine</td>
<td>7</td>
<td>98 ± 2.1</td>
<td>31 ± 0.49</td>
<td>27 ± 17</td>
</tr>
<tr>
<td>Thalamic</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Normal</td>
<td>5</td>
<td>109 ± 2.5</td>
<td>35 ± 0.9</td>
<td>21 ± 13</td>
</tr>
<tr>
<td>Adrenalectomy</td>
<td>4</td>
<td>107 ± 4.4</td>
<td>34 ± 1.5</td>
<td>25 ± 14</td>
</tr>
<tr>
<td>Pontine</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Normal</td>
<td>6</td>
<td>109 ± 5.0</td>
<td>32 ± 0.6</td>
<td>27 ± 13</td>
</tr>
<tr>
<td>Adrenalectomy</td>
<td>5</td>
<td>101 ± 3.4</td>
<td>33 ± 0.5</td>
<td>31 ± 13</td>
</tr>
<tr>
<td>Propranolol</td>
<td>4</td>
<td>113 ± 1.6</td>
<td>32 ± 1.2</td>
<td>25 ± 16</td>
</tr>
<tr>
<td>Atropine</td>
<td>7</td>
<td>112 ± 3.2</td>
<td>32 ± 1.7</td>
<td>25 ± 14</td>
</tr>
<tr>
<td>De-efferented</td>
<td>6</td>
<td>95 ± 4.0</td>
<td>31 ± 1.5</td>
<td>32 ± 16</td>
</tr>
</tbody>
</table>

*Mean ± SE to indicate range of variation; see Methods for details of statistical analysis.
††SE obtained by analysis of variance and based on within-animal comparisons; see Methods for details of statistical analysis.
§§pH significantly higher than resting (air) value, and significantly different from response of normal group; P < 0.01.
¶pH significantly below resting (air) value; P < 0.01.
▼As above, but P = 0.05.
throughout ($P<0.001$), and rose to approximately 80% of resting at the end of hypoxia (Fig. 4). There was a transient elevation in arterial and right atrial pressures at the time of maximum bradycardia, but both these variables gradually returned to the resting value toward the end of hypoxia. In normal pontine animals, the arterial pressure response was similar to that of the other group, but the right atrial pressure did not change significantly; the heart rate response was biphasic with a small initial bradycardia ($P<0.02$) of 1 to 2 minutes duration followed by a rise to $126\pm2.3\%$ ($P<0.001$) which was maintained for the remainder of hypoxia. In adrenalectomized sham-operated (and to a lesser extent in adrenalectomized thalamic animals) hypoxia caused a faster and larger fall in arterial and right atrial pressure from the maximum than in the corresponding normal groups ($P<0.001$ in sham-operated group) as well as a complete return to resting in the heart rate. On the other hand, the response of adrenalectomized pontine animals to hypoxia was not significantly different from that of normal pontine animals.

The vagal and sympathetic components of the heart rate response to hypoxia were examined in sham-operated and pontine animals (Fig. 5). Propranolol did not modify the bradycardia seen within a minute of the onset of hypoxia, so it would seem that this early chronotropic effect of oxygen lack is mainly vagal.

Atropine greatly reduced the hypoxic bradycardia in the sham-operated animal; however, the heart rate remained below resting throughout hypoxia, indicating that
there is a maintained reduction of cardiac chronotropic sympathetic activity during exposure of these animals to oxygen lack.

In the atropinized pontine animal subjected to oxygen lack, the heart rate rose to 120 ± 2.5%, indicating that the tachycardia in the normal pontine animal was due to an increased sympathetic activity. In propranolol-treated pontine animals, the heart rate fell to 88 ± 2.1% (P = 0.01), indicating that the initial slight bradycardia was largely vagal. During the latter part of hypoxia the heart rate remained at 94 ± 0.8%, significantly lower than in de-efferented animals (P < 0.0001); this could be due either to a slight but persistently increased vagal activity or to some depression by propranolol (17).

**Portal Vascular Resistance.**—In normal sham-operated animals the portal vascular resistance rose to a maximum of 468 ± 25% after 1 minute of hypoxia, and then declined, reaching a value of 122 ± 11% (0.1 > P > 0.05) at the end of hypoxia (Fig. 6). This return toward resting values does not indicate an absence of autonomic constrictor effects near the end of hypoxia, since in the autonomically de-efferented animals the portal resistance fell to only 38 ± 5.6% of the resting value at the end of hypoxia presumably because of a direct, local peripheral vasodilatation. In the normal group the portal vascular resistance at the end of hypoxia was significantly higher (P < 0.0001) than in the de-efferented rabbits, indicating that an increased autonomic constrictor activity had offset the direct local peripheral vasodilatation (12).

Hypoxia in thalamic rabbits caused a smaller initial maximum rise in portal resistance (to 342 ± 27%, P = 0.01) than in the sham-operated rabbits, and the peak response was not reached till after 4 minutes, but from this time until the end of hypoxia the changes were closely similar and the mean vascular resistance values of thalamic and sham-operated animals (171 ± 9.7% and 181 ± 6.3% of resting) were approximately the same. In pontine animals the rise to the peak (242 ± 11%) was more gradual still, and the rise was less well maintained, with the mean vascular resistance (137 ± 3.7%) significantly lower than in the other groups (P < 0.01). In sham-operated and pontine rabbits, propranolol did not modify the portal vascular response.

Adrenalectomy strikingly reduced portal vasoconstriction during hypoxia in sham-operated and pontine animals (mean portal resistance 103 ± 4.0% and 93 ± 4.0%, respectively) but had little effect (mean 161 ± 6.5%) on that of the thalamic rabbits (Fig. 6). In the adrenalectomized thalamic animals the neural portal constrictor effect was thus significantly greater than in the other adrenalectomized preparations, while that of sham-operated animals was greater than in pontine animals (P = 0.05) only at the beginning of hypoxia. These results suggest that increased adrenal secretions contribute significantly to the portal...
CENTRAL CONTROL OF REFLEX AUTONOMIC RESPONSE

**FIGURE 6**

Top: Mean portal vascular resistance (percent of resting) during hypoxia and recovery in sham-operated, thalamic and pontine animals with adrenals intact (open circles = propranolol-treated rabbits). Bottom: Results from adrenalectomized preparations and de-efferented rabbits. Hypoxia induced between arrows. Number of animals in Table 1.

**FIGURE 7**

Mean renal vascular resistance (percent of resting) during hypoxia and recovery in normal sham-operated, thalamic, and pontine rabbits and in de-efferented animals. Hypoxia induced between arrows. Number of animals in Table 1.
Mean muscle vascular resistance (percent of resting) during hypoxia and recovery in A, normal sham-operated, thalamic, and pontine animals; B, propranolol-treated sham-operated and pontine rabbits; C, after adrenalectomy in sham-operated, thalamic and pontine animals, and also in de-efferented rabbits. Hypoxia induced between arrows. Number of animals in Table 1. Inset: Schematic illustration of relationship between muscle vascular resistance (increase; — decrease from resting) in response to increasing sympathetic constrictor nerve activity and rising epinephrine concentration (after Celander, ref. 20). Point S corresponds to maximum dilator effect of epinephrine in normal sham-operated and pontine animals, while point P or P1 corresponds to the late epinephrine effect in normal pontine animals as discussed in text.

Constrictor response to hypoxia in normal sham-operated and pontine animals, but play virtually no part in the hypoxic response of the normal thalamic preparation.

Renal Vascular Resistance.—The changes in renal vascular resistance in normal sham-operated animals followed a pattern similar to the changes in portal vascular resistance, (Fig. 7). In normal thalamic animals the initial component of the response was significantly smaller than in normal sham-operated rabbits (P < 0.01), but the rise in resistance during the latter part of hypoxia was more pronounced (P < 0.001). In normal pontine animals the rise in vascular resistance was significantly smaller than in the other two groups (P < 0.001), and this indicated a minimal renal autonomic effect since their resistance exceeded the value of autonomically de-efferented animals only at the beginning and end of hypoxia (P < 0.01). We did not study the renal effects following adrenalectomy (see Methods).

Muscle Vascular Resistance.—In normal sham-operated rabbits, this fell gradually during hypoxia and reached a steady-state value of about 50% of the resting value after 15 to 20 minutes of hypoxia (57 ± 2.8% of resting) (Fig. 8A). After propranolol, hypoxia caused a rise in vascular resistance to 170 ± 8.6%. This indicates that the normal dilator response results from a preponderance of beta-receptor dilator effects over alpha-receptor constrictor effects, the latter becoming unmasked after propranolol (9). In adrenalectomized sham-operated animals muscle vascular resistance also increased significantly, with a rise at the beginning and a secondary rise toward the end of hypoxia (mean 124 ± 4.6%, P < 0.01; Fig. 8C), indicating that neural constrictor effects predominated over any neural beta-receptor dilator effects (18). The main cause of beta-receptor effects during hypoxia is probably epinephrine (virtually the sole adrenal medullary hormone in the rabbit [19]) produced in relatively small amounts corresponding to point S in the inset to Figure 8 (20).

In normal pontine animals the vascular resistance fell after about 8 minutes to 52 ± 9.8% (more rapidly than in sham-operated animals) but subsequently rose again to 88 ± 6.2% at the end of hypoxia. After propranolol there was a small but significant rise in resistance to a mean of 112 ± 2.8% (P = 0.05), greater than the mean of 74 ± 3.5% in normal pontine animals without the drug (P < 0.001). Hypoxia caused only trivial changes in vascular resistance in adrenalectomized rabbits in this group; they differed little from those seen in autonomically de-efferented animals (mean adrenalectomized pontine and de-efferented, 95 ± 4.2% and 96 ± 5.0%, respectively). Such findings indi-
Central Control of Reflex Autonomic Response

Figure 9
Mean vascular resistance in ear skin (percent of resting) during hypoxia and recovery in normal (solid circles) and adrenalectomized (open circles) sham-operated thalamic and pontine rabbits, and in de-efferented animals. Hypoxia induced between arrows.

cate that the neural constrictor effects produced by oxygen lack in the pontine group were minimal.

In the normal pontine preparation the initial vasodilator response in muscle during hypoxia is probably due to increasing secretion of epinephrine, with the initial effects corresponding approximately to point S in the inset to Figure 8, while the latter effects correspond to point P. Because of the nonlinear nature of the dose-response curve, the late data could also be explained by diminution in epinephrine concentration corresponding to point P1, but this is unlikely in view of the response after propranolol and the progressive fall in pH (21).

In normal thalamic animals the vascular resistance in muscle increased significantly to a mean of 133 ± 3.1%, contrasting with the findings in the other normal groups. The response remained essentially unaltered after adrenalectomy (145 ± 6.5%).

Cutaneous Vascular Resistance.—In both normal sham-operated and the normal thalamic rabbits, there was marked and rapid decrease in the vascular resistance in the ear (Fig. 9), to levels significantly below that of autonomically de-efferented animals; this suggests that the fall was due to reduction in cutaneous vasoconstrictor tone (13). However, in the pontine group the vascular resistance increased significantly (P < 0.02) soon after the onset of hypoxia; thereafter it fell gradually to slightly below resting values. The response of sham-operated animals was diminished after adrenalectomy (P < 0.001), but that of thalamic and pontine animals was little affected.

Discussion

In pontine animals increased cardiac sympathetic neural activity and secretion of epinephrine occur during arterial hypoxia in response to changes in input arising from the arterial chemoreceptors and baroreceptors (7), and there is also a moderate increase in portal constrictor effect (Fig. 10). However, the bulbar centers themselves seem to contribute little to reflex cardio-inhibitory or cutaneous dilator effects or to constrictor effects in the vascular beds of muscle and kidney induced by hypoxia in the intact rabbit.

In the thalamic preparation there is little evidence of the secretion of adrenal catecholamines during hypoxia. This suggests that the thalamus and hypothalamus inhibit this effect of the pontomedullary centers. Diencephalic centers seem to mediate the marked reflex cardiac slowing and inhibition of cutaneous constrictor tone in the ear by hypoxia (13), and are the major source of increased vasoconstrictor effects in renal and muscle beds and also contribute to the rise in portal resistance. The nature of the cardioinhibitory
Schematic diagram illustrating approximate contribution of cardiovascular centers in pons and medulla, thalamus and hypothalamus, and cerebral hemispheres, to the autonomic effector response of the rabbit during severe arterial hypoxia. The magnitude of the afferent inputs is considered to remain approximately constant, apart from changes in stimulus-profile after adrenalectomy and propranolol discussed in the text; the suggested projections from arterial chemoreceptors, baroreceptors and lung inflation receptors were considered in reference 7. Solid, downward-flowing arrows indicate excitation of autonomic effector to which arrow is pointing; open arrows indicate inhibitory effect on underlying effector or center. Size of arrows is proportional to approximate magnitude of effect mediated through the particular centers.

The adrenal secretions are of major importance in the normal sham-operated rabbit's reflex response to hypoxia, both by increasing muscle blood flow and by markedly accentuating the rise in portal vascular resistance. In fact, their contribution to the latter seems out of all proportion to the likely secretion rate of epinephrine (9, 20), which can be estimated approximately from the data of Chalmers et al. (9), who reproduced similar vasodilator effects in muscle by intra-aortic infusion of epinephrine (0.04 to 0.08 μg/kg/min). Interpretation of the differences in findings between normal sham-operated animals with adrenals intact and adrenalectomized rabbits is complicated by differences in the input stimulus-profile. In the intact hypoxic rabbit, stimulation of arterial chemoreceptors in conjunction with changes in baroreceptor input reflexly induces the increase in neural constrictor effects in portal, renal and muscle beds, the rise of epinephrine secretion, as well as the cardioinhibitory effects and the reduction in cutaneous constrictor tone (7, 12, 13, 15, 22-24), while the lung inflation reflex tends to inhibit these primary reflex circulatory chemoreceptor effects through a projection to the cerebral hemispheres (7, 25, 26), with the net autonomic effects depending on the ratio of these two sets of inputs (7). Because of the respiratory alkalosis that develops in sham-operated adrenalectomized animals, the arterial chemoreceptor input must be somewhat smaller than in normal animals, but because of the unchanged increase in respiration the input from lung inflation receptors will remain unaltered, tending to inhibit more easily the primary reflex chemoreceptor effects. This may account for the smaller portal constrictor response, greater reduction in blood pressure, less marked cardioinhibitory effects and smaller cutaneous dilator response to hypoxia in

Cerebral Hemispheres

Thalamus and Hypothalamus

Pons and Medulla

Cardiac Nerves

Sympathetic Constrictor Nerves

Adrenal Medulla


Effects in the thalamic preparation has not been analyzed in the thalamic preparation, but we have assumed in Figure 10 that they are the same as in sham-operated animals in view of our previous findings during controlled ventilation (7).

With cerebral hemispheres intact, epinephrine secretion again increases (21), while the neural effects are closely similar to those in the thalamic preparation though their onset is more rapid in portal and renal beds. The increased epinephrine secretion could result from cortical suppression of the proposed diencephalic-bulbar inhibitory mechanism or may alternatively result from a direct cortical excitation of the bulbar centers. The pontomedullary heart rate centers probably do not contribute directly to the response of the normal sham-operated animal at this particu-

lar PaO₂, since vagal and sympathetic effects of the latter group are both cardioinhibitory. However, previous analysis has shown that both during milder (7) and more severe (12) hypoxia there is a component of increased sympathetic activity with a time course similar to that of the pontine animal.

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this group. On the other hand, in propranolol-treated animals although the degree of respiratory alkalosis is similar, the respiratory response to hypoxia is less, and the maintenance of a normal portal constrictor effect can probably be accounted for by the approximate constancy of ratio of chemoreceptor inputs to lung inflation inputs. It seems likely because of the changes in the stimulus-profile after adrenalectomy that the neural effects in adrenalectomized animals will be smaller than the "true" neural effects in normal animals with adrenals intact.

The adrenal catecholamines probably influence the autonomic responses in the intact animal through their peripheral effects on alpha and beta receptors in vessels, and also through their direct (27) and systemic metabolic effects (i.e., circulating H⁺) on arterial chemoreceptors. In addition, adrenal steroids are probably secreted during hypoxic "stress" and may enhance vascular sensitivity to catecholamines (28). The present experiments leave undecided the relative importance of the above factors, but the action of the adrenal catecholamines appears to be much greater in the framework of the whole circulatory control loop of the unanesthetized animal, than in more isolated preparations. Their importance in the present experiments is in marked contrast to the rather slight role ascribed to them in current physiological teaching (4, 20), much of which is based on experiments under anesthesia where some of the above mechanisms may become relatively depressed.

The responses of the various preparations serve as illustrative models of alternative mechanisms of circulatory control during pronounced impairment of the body oxygen supply. We can view the three autonomic responses against that of the autonomically de-efferented group in which peripheral autoregulation is adequate to prevent excessive formation of H⁺ ion (Table 3), but where the portal vasodilation caused by the local effects of hypoxia takes an excessive fraction of the cardiac output at the expense of the blood flow to the brain and heart (15). In the hypoxic pontine preparation, increased cardiac sympathetic activity contributes to the rise in cardiac output (7), and high rates of epinephrine secretion provide some relatively nonselective peripheral vasomotor control. The price for the high rates of epinephrine secretion is rapid metabolic production of H⁺ ions, which must limit long-term survival, particularly in association with the increased cardiac oxygen demand. In the thalamic preparation epinephrine secretion is inhibited, cardiac work and oxygen demand are reduced, and marked neurally mediated constriction produces redistribution of peripheral blood flow to the brain and heart. There is still considerable metabolic production of H⁺ despite the virtual absence of epinephrine secretion, probably related to the relatively higher anaerobic metabolism due to the restricted muscle perfusion. The normal animal with intact cerebral hemispheres has increased muscle blood flow again seemingly caused by increased secretion of epinephrine but otherwise the neural diencephalic mechanisms seem to operate. Such a control system redistributes blood flow optimally so that cardiac work and metabolic production of H⁺ ions are minimal and may favor longer survival during hypoxia.

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References


Bulbar and Suprabulbar Control of the Cardiovascular Autonomic Effects during Arterial Hypoxia in the Rabbit

JOHN B. Uther, STEPHEN N. HUNYOR, JOHN SHAW and PAUL I. KORNER

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