Equine Exercise Physiology 3: 1–8, 1991

Respiratory Muscle Perfusion during Strenuous Exercise

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ABSTRACT. The present study was carried out on 7 healthy ponies to examine blood flow in all respiratory muscles during strenuous exercise. Blood flow was ascertained using 15 μm diameter, radionuclide labeled microspheres injected into the left ventricle. Measurements were made during rest, maximal exercise (32 km h^{-1}), and at third and ninth min of exhaustive exercise (22 km h^{-1}) lasting 10 min. The data revealed that with each exercise bout, blood flow increased in all inspiratory as well as expiratory muscles. Among the inspiratory muscles, the costal diaphragm received the highest perfusion followed first by the crural diaphragm and serratus ventralis, and then by the external intercostal, serratus dorsalis, and scalenus muscles. Among the expiratory muscles, transverse thoracis, and the internal oblique abdominis had the highest flow followed by the transverse, rectus and external oblique abdominis muscles. In all muscles, perfusion was greater during maximal exercise as compared with exhaustive exercise. Perfusion in all respiratory muscles was similar for third and ninth min of exhaustive exercise. The latter was attended by a 3.9°C rise in core temperature, which necessitated cutaneous vasodilation and profuse sweating. Concomitantly, cardiac output rose and renal vasoconstriction occurred so that muscular perfusion was maintained at its proper level. It was concluded that: (1) all respiratory muscles are activated during exertion and their metabolic O_2 needs are quite diverse during exercise and (2) that in exhaustive exercise, respiratory muscle perfusion was well preserved in the face of marked cutaneous vasodilatation necessitated by the rising thermal burden.

Key words: Inspiratory muscles; expiratory muscles; blood flow; cardiac output; exercise; horses.

INTRODUCTION

Despite early recognition that metabolic needs of respiratory muscles during exercise may be substantial,[1,2,15] only recently has attention been focused upon examining how these demands are met. The impetus for this effort has primarily gathered from reports of inspiratory muscle fatigue in people following high intensity short-term as well as prolonged exertion,[3,8] and the suggestion that there may be competition between working limb muscles and the respiratory muscles for available cardiac output.[1,2] Among inspiratory muscles, the dominant role of diaphragm is well known; hence, it received most attention[7,9–13,16] while other muscles were largely ignored. There is also an increasing realization that exhalation is not a completely passive process,[4–6] as once believed; yet no data are available in any species examining metabolic requirements of expiratory muscles in exercise. The primary objective of this study, was to compare blood flow in all inspiratory and expiratory muscles[4–6] during short-term, strenuous exercise.

METHODS

Experiments were carried out on 7 healthy ponies weighing 206 ± 13 kg. They were housed in an air-conditioned building

(~ 20°C) and were fed a ration comprised of alfalfa hay and oats. Water was provided *ad libitum*. All ponies had been dewormed and inoculated with tetanus toxoid previously. They were familiarized with personnel, laboratory surroundings and with running on a motor driven treadmill.

Experimental protocol

Hemodynamic measurements were made during steady-state conditions. The existence of a steady state was judged based on the stability of heart rate (HR), and arterial and ventricular pressures. Each animal was studied during the following three conditions:

1. *Rest* (Baseline): The data were obtained from ponies standing quietly on the treadmill when heart rate and arterial and ventricular pressures had been stable for at least 20 to 25 min.

2. *Maximal exercise*: The animals were exercised for 4 min at maximal HR achieved at a treadmill speed setting of 32 km h^{-1}. The treadmill had a fixed grade of 7%, and the onset of exercise was rapid. Hemodynamic measurements were made in the third and fourth min of exercise under steady-state conditions. A rest period of 120 min followed to permit return of cardiorespiratory variables to control values.

3. *Exhaustive exercise*: Ponies were exercised for 10 min at a treadmill speed of 22 km h^{-1} + 7% grade, preselected to achieve a heart rate of ~ 190 beats min^{-1}. The onset of exercise was rapid. Heart rate, and arterial and ventricular pressures were continuously monitored for the duration of exercise, but tissue blood flow was determined only in the 3–4th and 9–10th min of exercise during steady-state conditions. A rest period of 120 min followed.

Procedures for hemodynamic study

The methodology used has been reported previously.[9–14,16] A brief description is given here. On the day of hemodynamic study, the left common carotid artery and the jugular vein were exposed in the mid-cervical region after local infiltration of 2% lidocaine HCl. A fluid filled cardiac catheter was advanced via the jugular vein into the main pulmonary artery. Fluid filled catheters were positioned into the left ventricle and the thoracic aorta via the carotid artery. The left ventricular catheter was used for injecting radionuclide labeled 15 μm diameter microspheres while reference blood was withdrawn from the thoracic aorta. Another cardiac catheter with dual tip-micromanometers was also advanced via the carotid artery, and its pressure sensors were positioned into the left ventricle and the aortic root. Heart rate was determined from the phasic pressure pulse in the left ventricle and/or aorta.

Tissue blood flow was determined using 15 to 18 million radionuclide labeled (^{141}Ce, ^{51}Cr, ^{85}Sr, ^{46}Sc) 15 μm diameter microspheres injected into the left ventricle while reference blood was being withdrawn at a constant rate of 28 ml min^{-1} from the thoracic aorta. Lack of aggregation of microspheres was verified before every injection by microscopic examination of a drop of the vigorously agitated, ultrasonicated suspension. Adequate mixing of microspheres with blood was demonstrated by similarity of left and right kidney blood flow values.

Upon completion of the experiment, ponies were anesthetized with intravenous thiamylal sodium and killed by exsanguination. Both kidneys, diaphragm, serratus dorsalis, serratus ventralis, scaleni, external and internal intercostal muscles, transverse thoracis (triangularis sterni), external and internal oblique abdominis, transverse abdominis and rectus abdominis muscles were removed carefully. After weighing, these tissues were carbonized at 300°C. The radioactive carbon was counted along with reference blood samples and tissue blood flow computed.[9–11,14,16] Tissue vascular resistance was calculated as mean aortic pressure (mmHg) divided by blood flow (ml min^{-1} g^{-1}). Total peripheral resistance was calculated as mean aortic pressure (mmHg) divided by cardiac output in ml min^{-1} kg^{-1}. These values are considered to represent crude estimates only

Table 1. *Hemodynamic variables at rest and during exercise*

All values are mean ± SEM. The * denotes significant differences from rest, † denotes significant differences from maximal exercise and § denotes significant differences from values at third min of exhaustive exercise

Variables	Rest	Maximal exercise	Exhaustive exercise: Third min	Exhaustive exercise: Ninth min
Heart rate	55	218*	190*†	204*†
(beats min^{-1})	±3	±4	±6	±7
Aortic systolic	159	222*	216*	199*
pressure (mmHg)	±8	±6	±12	±8
Mean aortic	125	170*	169*	156*
pressure (mmHg)	±5	±6	±8	±4
Aortic diastolic	97	128*	124*	118*
pressure (mmHg)	±5	±7	±7	±5
Cardiac output	96	730*	478*†	710*§
(ml min^{-1} kg^{-1})	±11	±78	±53	±90
Total peripheral vascular	1.42	0.25*	0.40*†	0.24*§
resistance (mmHg/ml min^{-1} kg^{-1})	±0.18	±0.09	±0.06	±0.03
Blood flow to left kidney	669	130*	604†	244*†§
(ml min^{-1} 100 g^{-1})	±72	±109	±129	±121
Blood flow to right kidney	635	118*	562†	211*†§
(ml min^{-1} 100 g^{-1})	±66	±99	±130	±113

because the resistance calculations did not take into account the back pressure opposing perfusion.

Statistical analysis

The data were subjected to two-way analysis of variance followed by Newman Keul's multiple range test[19] to determine significant effects of various work loads at $p<0.05$. For each exercise intensity, tissue blood flow and vascular resistance for various muscles were also compared using analysis of variance procedures.[18] Data are presented as mean ± SEM.

RESULTS

Heart rate. Significant increment in HR occurred with exercise; the values achieved during exhaustive exercise were, however, less than that for maximal exercise (Table 1).

Phasic and mean aortic pressure. Marked rise in aortic pressure occurred with exercise, but statistically significant differences between maximal exercise and exhaustive exercise did not occur (Table 1).

Cardiac output and stroke volume. Maximal exercise resulted in a 7.6-fold rise in cardiac output (Table 1). At third min of exhaustive exercise, cardiac output was 5-fold its resting value, but by ninth min it had reached a value similar to maximal exercise (Table 1).

With maximal exercise, stroke volume increased significantly to 3.4 ml $beat^{-1}$ kg^{-1} from a resting value of 1.75 ml $beat^{-1}kg^{-1}$. At third min of exhaustive exercise (2.52 ml $beat^{-1}$ kg^{-1}) it was less than during maximal exercise, but by ninth min (3.48 ml $beat^{-1}$ kg^{-1}) it was similar to the maximal exercise value.

Systemic vascular resistance. A marked decrease in total peripheral resistance occurred with maximal exercise. At third min of exhaustive exercise, total peripheral resistance was 28% of the resting value, but by ninth

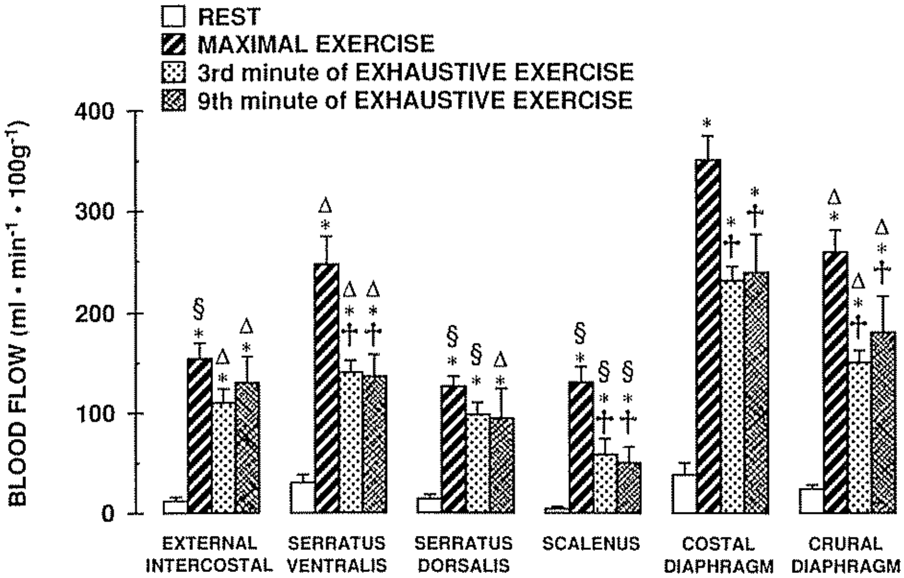

Fig 1. Inspiratory muscle perfusion during exercise. * = Significantly different from rest. † = Significantly different from maximal exercise. Δ = Significantly different from corresponding costal diaphragmatic flow value. § = Significantly different from corresponding blood flow values for the costal diaphragm, crural diaphragm and serratus ventralis muscles. Blood flow values at third and ninth min of exhaustive exercise were similar in all inspiratory muscles.

min it had decreased to 16.9% of the resting value (Table 1) and was not different from maximal exercise.

Renal blood flow Values for left and right kidneys were similar at each step of the protocol. During maximal exercise, renal perfusion decreased to 19% of its resting value. At third min of exhaustive exercise, renal blood flow was similar to resting value, but by ninth min it was only 35% of the resting value.

Inspiratory muscle blood flow (Fig 1) and vascular resistance (Table 2). Both exercise bouts resulted in highly significant increments in blood flow to all inspiratory muscles as marked vasodilation occurred. Highest perfusion was recorded in the costal diaphragm followed first by the crural diaphragm and the serratus ventralis, and then by the external intercostal, serratus dorsalis, and scalenus muscles. In all inspiratory muscles, largest perfusion values occurred during maximal exercise. Interestingly, values for third and ninth min of exhaustive exercise were similar in all inspiratory muscles. Collectively, the inspiratory muscles received 6.17±1.28, 8.44±1.05, 7.85±0.77 and 5.05±0.42% of the cardiac output at rest, maximal exercise, and third and ninth minutes of exhaustive exercise.

Perfusion (Fig. 2) and vascular resistance (Table 2) in expiratory muscles. With exercise, blood flow in all expiratory muscles increased significantly as vascular resistance decreased markedly; highest perfusion occurring in the transverse thoracis and the internal oblique abdominis muscles. Again, highest perfusion values for expiratory muscles were observed during maximal exercise. Blood flow values for third and ninth min of exhaustive exercise also remained similar in all expiratory muscles examined. Collectively, the expiratory muscles received 3.75±0.93, 6.35±0.28, 6.90±0.28, 4.29±0.17% of the cardiac output at rest, maximal exercise, and third and ninth minutes of exhaustive exercise.

In comparison with costal diaphragm, the

Table 2. *Vascular resistance (mmHg/ml^{-1} min^{-1} g^{-1}) in various respiratory muscles at rest and during exertion*

All values are mean ± SEM. The * denotes significant difference from rest, and the † denotes significant difference from maximal exercise. Values for third and ninth min of exhaustive exercise remained similar in all respiratory muscles

Variables	Rest	Maximal exercise	Exhaustive exercise Third min	Exhaustive exercise Ninth min
A. *Inspiratory muscles*				
Diaphragm	751	50*	74*†	75*†
	±283	±4	±3	±14
External intercostal	1 294	119*	167*†	147*†
	±212	±12	±19	±27
Serratus dorsalis	1 622	143*	195*	264*†
	±454	±15	±38	±71
Serratus ventralis	676	75*	126*†	155*†
	±175	±10	±13	±51
Scalenus	5 208	162*	438*†	813*†
	±1 292	±41	±119	±437
B. *Expiratory muscles*				
Transverse thoracis	685	73*	118*†	121*†
	±183	±5	±8	±20
Internal intercostal	1 455	112*	149*†	144*†
	±500	±16	±30	±26
External oblique abdominis	2 146	171*	252*†	291*
	±842	±25	±50	±102
Internal oblique abdominis	1 829	75*	111*†	114*†
	±771	±7	±21	±22
Transverse abdominis	1 662	164*	288*†	288*†
	±334	±26	±64	±74
Rectus abdominis	6 090	136*	212*†	277*†
	±3 917	±18	±26	±76

internal oblique abdominis and transverse thoracis received less blood flow (per unit weight basis), but their perfusion values during maximal, as well as exhaustive exercise, compared favorably with serratus ventralis and crural diaphragm (Fig. 1). Exercise blood flow values for external and internal intercostal muscles were also similar.

Pulmonary artery blood temperature. From its resting value of 37.9±0.1°C, it increased to 40.4±0.2°C at third min of maximal exercise. During third and ninth min of exhaustive exercise, respective values were 39.4±0.2 and 41.8±0.3°C. Despite the ambient temperature being held at 15–16°C, the ponies sweated profusely.

DISCUSSION

The major findings of this study are: (1) all inspiratory and expiratory muscles are activated during exercise as demonstrated by significant increments in their perfusion; (2) metabolic O_2 requirements of individual respiratory muscles during exercise are diverse as demonstrated by the marked heterogene-

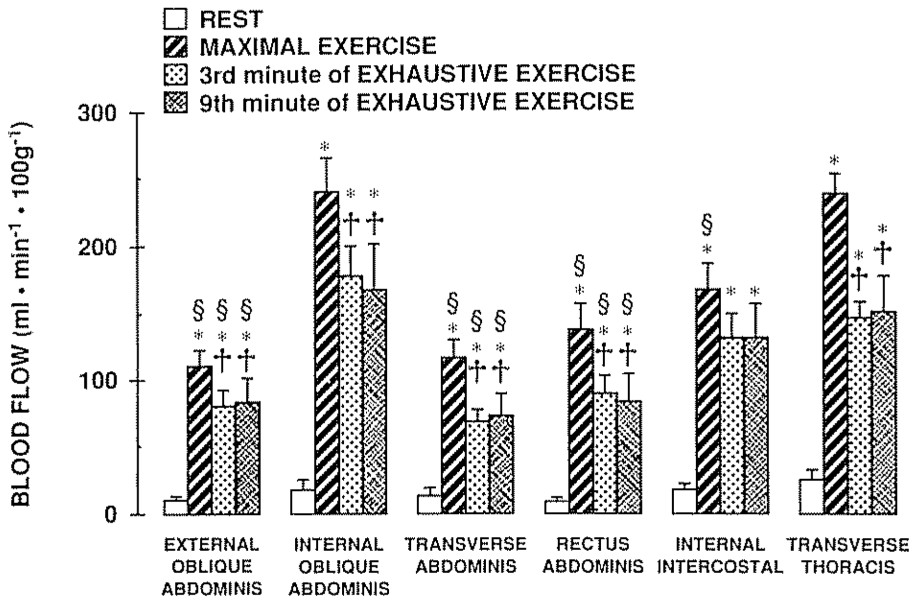

Fig. 2. Expiratory muscle perfusion during exercise. * = Significantly different from rest. † = Significantly different from maximal exercise. § = Significantly different from corresponding blood flow values in the internal oblique abdominis, and the transverse thoracis muscles. Blood flow values for the third and ninth min of exhaustive exercise were similar in all expiratory muscles.

ity of their perfusion; (3) the respiratory muscles comprised 5.5±0.1% of the body weight, and they received 9.92±2.08%, 14.8±1.2%, 14.8±1.15% and 9.33±1.1% of the cardiac output during rest, maximal exercise, and the third and ninth minutes of exhaustive exercise, respectively; (4) it was demonstrated in exhaustive exercise of 10 min duration that respiratory muscle perfusion was well preserved in the face of marked cutaneous vasodilatation necessitated by the rising thermal burden.

Muscles discretely involved in respiration in resting or exercising horses have not been identified. Confronted with lack of this vital information, it was decided to study those muscles which have been reported to be involved in respiration in man and the dog.[4–6] Even in these species, the precise contribution of some of these muscles to inspiration and exhalation processes is not precisely identified.[6] A note of caution is therefore, warranted because it is not certain that the alterations in blood flow of various muscles of exercised ponies (Figs. 1 and 2) were due entirely to their work related to respiration.

Inspiratory muscle perfusion in exercise

It is well accepted that the magnitude of blood flow in various tissues closely mimics their metabolic O_2 needs[1,2] as determined by the invoked work effort. The data in Fig. 1 indicate that all inspiratory muscles increased their work effort considerably during exercise. Although it has been demonstrated previously that costal diaphragmatic blood flow in exercising ponies exceeded that in the crural diaphragm,[9] it is being reported for the first time that costal diaphragmatic perfusion exceeds that of every other inspiratory and expiratory muscle. Among the remaining inspiratory muscles, the serratus ventralis not only had the greatest bulk, but also received higher blood flow than all others, including the external intercostal muscles. This was surprising in that external intercostal muscles are thought to have an important role in assisting the inspiratory effort of the diaphragm.[6] At present, the lack of similar studies in other species prevents comparison with the horse. In addition, whereas O_2 and lactate extraction by the equine diaphragm has been studied,[12,13] such knowledge regarding other respiratory muscles awaits further work.

Expiratory muscle perfusion in exercise

In anesthetized dogs, the contribution of the transverse thoracis and transverse abdominis to the act of exhalation has been demonstrated.[5] In keeping with these observations, a significant contribution of the transverse thoracis to exhalation in exercising ponies may be noted since its blood flow increased markedly. Its bulk in ponies is, however, much smaller relative to other expiratory muscles. In discord with previous observations on dogs,[4] among the abdominal muscles of exercising ponies blood flow was found to be greatest in the internal oblique abdominis. Although data in Fig. 2 indicate that all expiratory muscles were active in exercising ponies, their individual contribution to the act of exhalation awaits further work. Perfusion in the internal intercostal muscles of exercising ponies remained similar to the external intercostal muscles, thereby suggesting a similar work effort of the two sets.

Respiratory muscle perfusion at third and ninth min of exhaustive exercise

Thermal burden rose markedly with exhaustive exercise as evidenced by 3.9°C rise in pulmonary artery blood temperature, and the ponies sweated profusely. The total peripheral resistance decreased by 40% between the third and ninth min of exercise indicating extensive cutaneous vasodilatation. It has been suggested that when core temperature rises markedly in exercise, there may be competition between the cutaneous and working skeletal muscle vascular beds for available cardiac output.[1,2,18] The pony's response to the difficulty of providing for cutaneous vasodilator need (i.e. for thermoregulation) at a time of high metabolic O_2 requirements of working muscles was twofold. First, with extensive cutaneous vasodilatation as total peripheral resistance (i.e. the afterload) fell, the ventricles increased their stroke volume by 38.1%. This and the rise in HR resulted in a 48% increase in cardiac output. The additional cardiac output could be employed for thermoregulatory needs without the necessity to curtail/divert blood flow from working muscles. Afterload dependence of stroke volume at constant preload and contractility is well known.[17] Secondly, in accordance with previous observations,[16] renal blood flow at the third min of exhaustive exercise was similar to resting value, but it decreased markedly at the ninth min indicating renal vasoconstriction. Changes in renal blood flow of exercising ponies generally tend to parallel those in splanchnic organs.[11] Thus, in the extenuating circumstances of high metabolic O_2 needs of working muscles and rising core temperature at the ninth min of exercise, the ponies diverted blood flow away from renal, and possibly splanchnic, vascular beds so that muscle blood flow would not suffer. Sweating induced fluid loss may have necessitated the renal, and possibly splanchnic, vasoconstriction at the ninth min of exercise. In the present study, the duration of exhaustive exercise was only 10 min and it is not known if these findings would extend to exercise of a longer duration.

The events described in these ponies are somewhat different from findings in humans performing prolonged exercise.[18] First, in exercising humans, the renal and splanchic perfusion decrease in proportion to exercise intensity,[18] in contrast with ponies where renal blood flow is unaffected with short-term, submaximal exercise.[16] This difference is apparently related to the erythrocyte reservoir function of the equine spleen.[11] Secondly, differences exist between humans and horses in relation to the changes in cardiac output as exercise duration is prolonged. In humans, as thermoregulatory demand causes skin blood flow (and volume) to increase, the central blood volume shrinks and cardiac output can only be maintained by augmenting HR.[18] The inability to increase stroke volume in humans during prolonged submaximal exercise[18] is in striking contrast with the findings of this study. This probably reflects the differences in the distribution of blood volume in humans (i.e. bipeds) and quadrupeds. Whereas in humans, nearly 70% of the blood volume is situated below the level of the heart, in quadrupeds 70% of their blood volume is at or above the level of the heart.[18] This difference can have a profound influence on venous return and cardiac filling in the two species.[17,18] In prolonged exercise, sweating induced fluid loss may confound the problem of shrinking central blood volume[18] because offsetting features viz., splenic blood volume reservoir and the renal and splanchnic vasoconstriction, are either lacking or have already been invoked. These factors indicate that ponies are better adapted to perform prolonged exercise than humans.

In conclusion, this study demonstrates that all inspiratory and expiratory muscles are activated during exercise and that their individual metabolic O_2 needs are quite diverse. In exhaustive exercise of 10 min duration, respiratory muscle perfusion is well preserved in the face of marked cutaneous vasodilatation necessitated by the rising thermal burden.

ACKNOWLEDGEMENTS

The excellent technical assistance of David Nganwa, Rand Gustafson, Susan Mende, and Shanon Turner is gratefully acknowledged. This work was supported in part by a Grant-in-Aid from the American Heart Association, Illinois Affiliate.

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