PHRENIC BLOOD FLOW IN CATS WITH A CLOSED ABDOMINAL CAVITY

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In many investigations the blood supply to the respiratory muscles has been studied with the aid of radioactive microspheres [9, 10, 12]. By means of this method quantitative data on the blood supply of a particular organ at a particular time of the experiment can be obtained, but it is impossible to judge the dynamics of the process. In some studied the blood flow to the diaphragm has been studied with the aid of cannulative blood flow transducers [7, 13, 15] or after opening of the abdominal cavity [7, 11, 14]. The blood flow along the intercostal arteries was studied previously in the writers' laboratory [6]. In our view it is important to study the blood flow to the diaphragm by an up to date method in which, besides obtaining quantitative data, it would also be possible to study the dynamics of the blood supply and also the true character of the pulse flow in the phrenic artery under different conditions.

The aim of this investigation was to study the linear and volume velocity of the blood flow in the phrenic artery (PA), the resistance of the vascular bed of PA, and the ratio of the changes in blood flow and resistance and the phases of the respiratory cycle and changes in the systemic arterial pressure (BP) during exposure to different factors.

EXPERIMENTAL METHOD

In acute experiments on 22 male and female cats weighing 2-4 kg, and under pentobarbital anesthesia (40-50 mg/kg, intraperitoneally), the linear and volume velocity of the blood flow in the left PA was studied by an ultrasonic method [1]. Measurements were made with transducers of bandage type with a slit 2 mm long and an internal diameter of 0.4-0.6 mm. The sensitive element consisted of miniature piezoelectric crystals with an area of 0.5 mm^2, working at a frequency of 26 mHz. The transducers were calibrated in blood flow volume velocity units. The linear blood flow velocity was determined from the value of the Doppler frequencies. BP was recorded in the femoral artery by means of a semiconductor micromanometer [2]. Values of the blood flow along PA and of the blood pressure were led into an analog computer, which calculated the resistance of the vascular bed of PA during the process as the quotient obtained by dividing mean values of the systemic BP by the mean volume velocity of the blood flow along PA [mm Hg/(ml/min)] The phrenic artery was approached through a midline incision in the abdominal wall. Without extracting the internal organs, they were moved into the right half of the abdominal cavity with the aid of towels soaked in physiological saline, so that the left kidney and adrenal were in the field of vision. With these guides the left PA was found, arising either from the celiac artery (in 11 experiments) or from the adrenal artery (in 11 experiments). PA was freed from connective tissue and fat and an ultrasonic transducer applied to it; the leads from the transducer were sutured to muscles of the dorsal wall of the abdominal cavity. The abdominal cavity was then sutured in layers. The lower part of the thorax was surrounded by a wide rubber band to which the strain-gauge transducer for recording respiratory excursions was fixed. Changes in blood flow were analyzed in different phases of the respiratory cycle, during quiet and more active breathing, caused by various procedures: changes in the composition of the gas medium, asphyxia; or administration of biologically active substances. The phrenic blood flow in cats has not previously been studied. Cats were chosen as the test object for comparison with the results of investigations which were carried out previously also on cats, but on different vascular regions: the pulmonary vessels [3], coronary arteries [4], and bronchial arteries [5].

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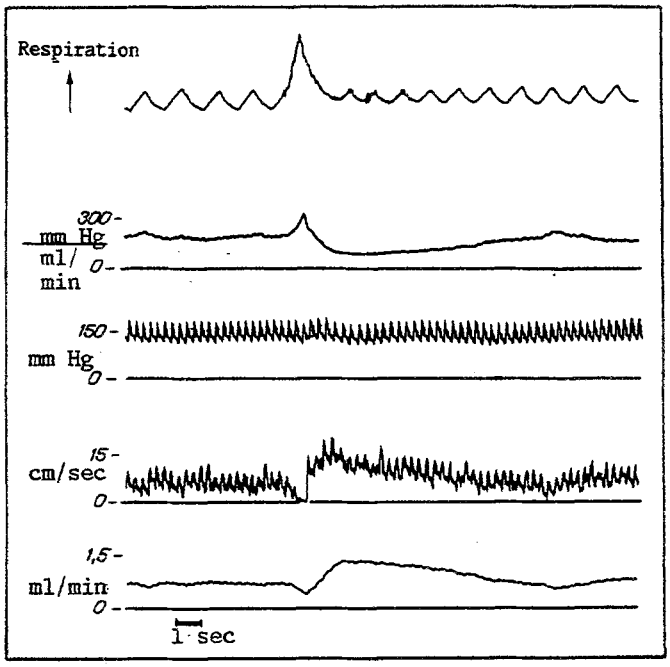

Fig. 1. Changes in resistance of vascular bed of phrenic artery and blood flow along phrenic artery during quiet and deep breathing. From top to bottom: respiration, resistance of vascular bed of phrenic artery, blood pressure in femoral artery, linear velocity of blood flow in phrenic artery, volume velocity of blood flow in phrenic artery. Here and in Figs. 2 and 3: thin lines beneath each curve denote zero levels.

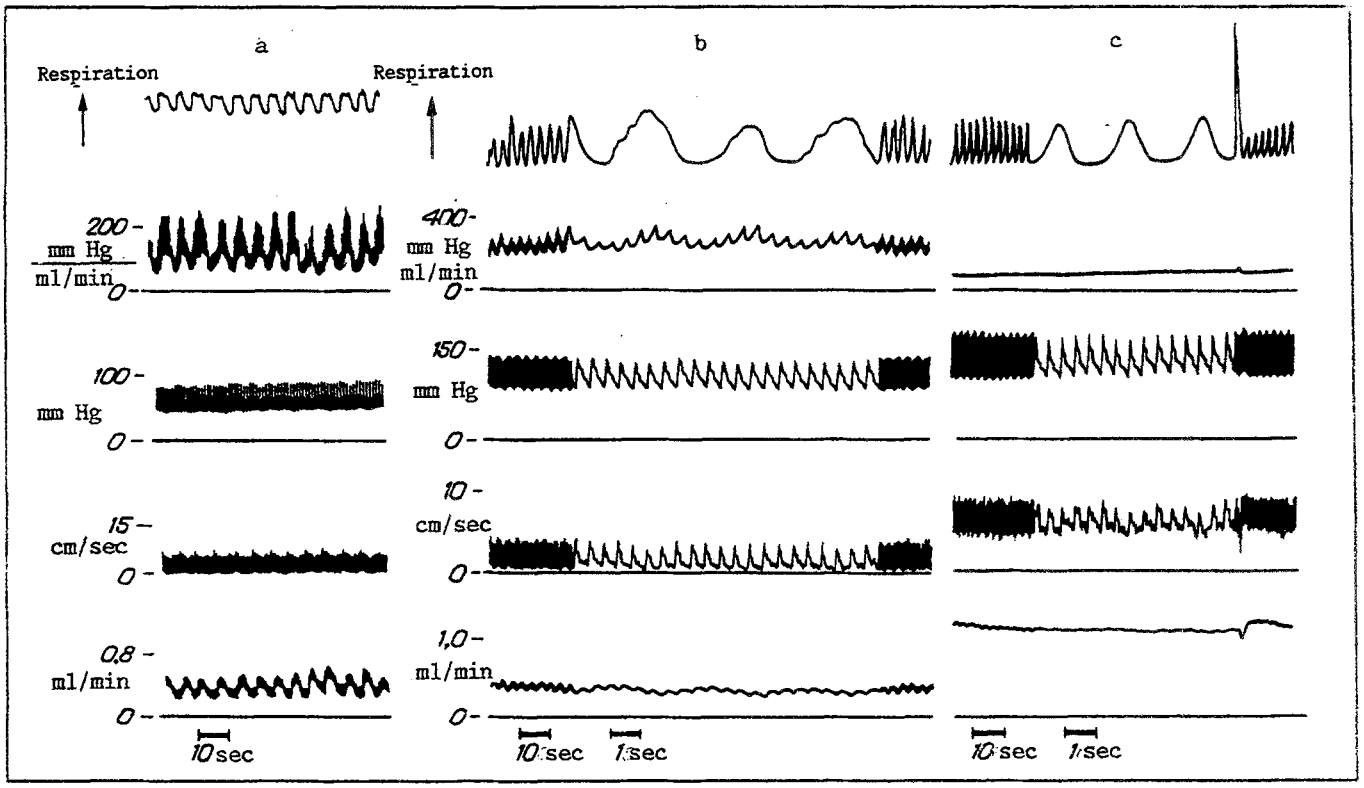

Fig. 2. Changes in resistance of vascular bed of phrenic artery and blood flow along phrenic artery during respiratory cycle. a) At a time of low systemic arterial pressure, b) of high arterial pressure and during inhalation of air, c) during inhalation of gas mixture containing 5% O_2 (in the same experiment). Significance of curves the same as in Fig. 1.

EXPERIMENTAL RESULTS

The volume velocity of the blood flow along PA averaged 0.39 ml/min (with fluctuations from 0.1 to 1.1 ml/min in different animals, most frequently — in 14 experiments – from 0.2 to 0.65 ml/min). These values are comparable with those obtained on dogs, namely about 3 ml/min, with body weight 8-10 times greater than the body weight of cats [11, 14]. The linear velocity of the blood flow in PA averaged 4.8 cm/sec (from 2 to 10.5 cm/sec). Incidentally, the values of the linear velocity of the blood flow in PA which we found are very low compared with the values which we observed in other vessels: 50-60 cm/sec in the pulmonary arteries, 30-40 cm/sec in the pulmonary veins [3], 20-25 cm/sec in the coronary arteries [4], and 10-20 cm/sec in the bronchial arteries [5]. The resistance of the vascular bed of PA averaged 310 mm Hg/(ml/min) [from 180 to 500 mm Hg/(ml/min)]. These values are much higher than those which we obtained previously in other vascular regions. The absolute values of the blood flow velocity were independent of the animal's body weight: in larger animals low values could be observed, and higher values of the volume and linear blood flow along PA in smaller animals. These parameters were determined by a greater degree by the animal's functional state at the beginning of the experiment and, in particular, by the initial systemic BP and the initial vascular resistance of PA.

The blood flow to the respiratory muscles is determined by relations between several influences. During muscular contraction with constant perfusion pressure the blood flow in the muscle depends on the action of two opposing factors: lowering of the vascular resistance, which increases the blood flow in accordance with the metabolic needs, and the intramuscular tension, which compresses the blood vessels and restricts the blood flow. During the phase of muscle relaxation the blood flow is determined by the vascular resistance and the level of the perfusion pressure. These regular principles hold good for skeletal muscle, and they apply equally well to the respiratory muscles [13-15].

The blood flow in PA has a distinct pulsatile character. Besides the constant component and the pulsatile fluctuations of blood flow, changes synchronized with phases of the respiratory cycle were sufficiently well marked: during inspiration (contraction of the diaphragm) the blood flow falls, during expiration it rises. Changes in the vascular resistance of PA at different phases of the respiratory cycle are opposite in direction to changes in the blood flow. These relationships are manifested particularly clearly during spontaneous deep inspiration (Fig. 1). We analyzed changes in vascular resistance and blood flow along PA in different phases of the respiratory cycle and expressed the value of the blood flow during inspiration as a percentage of blood flow during expiration, taken as 100%. During quiet breathing values of the linear blood flow in the phase of inspiration averaged 73.5%. These values showed considerable scatter – from 44 to 98%. In the majority of experiments the linear velocity of the blood flow during inspiration varied between 80 and 86% of the blood flow in the expiration phase. Values of the volume velocity of the blood flow during inspiration averaged 79% of the blood flow in the expiration phase, with fluctuations from 62.5 to 90% (most often from 80 to 90%). Analysis of individual experiments showed that the most marked "respiratory waves" of the volume blood flow and resistance of the vascular bed of PA were observed usually in animals with an initially low systemic BP (Fig. 2a). At a low BP, less tension of the muscle was required to compress the vessels and to stop or reduce the blood flow at inspiration [13]. An appreciable fall of blood flow and increase of resistance of the vascular bed of PA during inspiration could also be observed in experiments in which the initial level of resistance of the vascular bed of PA was increased; usually in these experiments the constant component of the blood flow on the curve of the pulsatile blood flow was ill defined or absent.

During forced breathing, brought about in different ways (hypoxia, hypercapnia) the linear and volume velocity of the blood flow was increased and the vascular resistance of PA reduced. The respiratory waves of the blood flow also were increased (Fig. 2a, b). Meanwhile the ratio between values of the blood flow in the phase of inspiration and expiration did not change significantly. The linear velocity of the blood flow under these conditions in the phase of inspiration averaged 69.9% of the blood flow in the expiration phase (with fluctuations from 48 to 80%). The volume velocity of the blood flow in the phase of inspiration averaged 82.4% of its value in the expiration phase (from 62 to 95%). Thus contractions of the diaphragm have a definite influence on resistance of the vascular bed of PA and of the blood flow along PA. The ratios of the values of the blood flow in the phase of inspiration and expiration did not differ significantly during quiet and forced breathing.

PA possesses large reserves for increasing the blood flow. With low initial values the resistance of the vascular bed of PA during forced breathing may be reduced, but the blood flow may be increased by many times. It has been shown that a four-fivefold increase in pulmonary ventilation is associated with a ten-twentyfold increase in the blood supply to the respiratory muscles [12]. In the present experiments, during short-term asphyxia (under 1 min) the resistance of the vascular bed of PA decreased, but the blood flow increased considerably, mainly after the ending of asphyxia, when it was aimed at

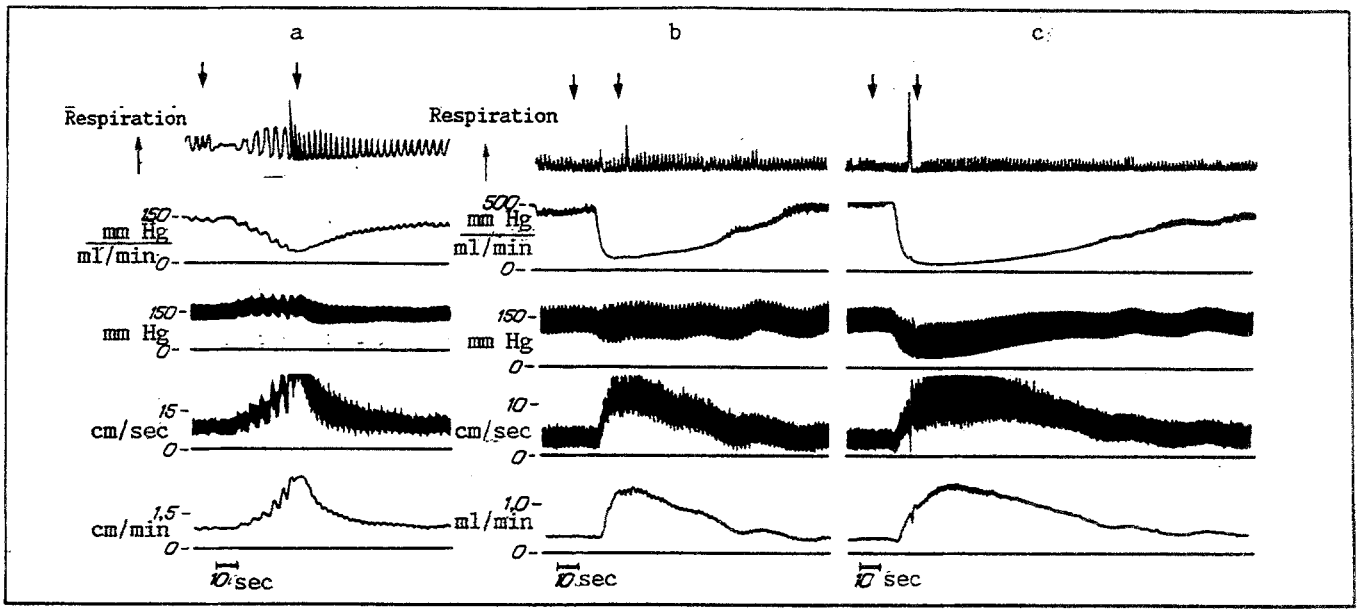

Fig. 3. Changes in resistance of vascular bed of phrenic artery and blood flow along phrenic artery: a) during asphyxia, b) during intravenous injection of adrenalin ($1 \cdot 10^{-8}$ g), c) during intravenous injection of histamine ($1 \cdot 10^{-8}$ g/kg). Significance of curves the same as in Fig. 1. Arrows indicate beginning and end of asphyxia (a), and injection of adrenalin or histamine (b, c).

abolishing the oxygen debt. Under these circumstances the linear velocity of the blood flow increased by 2-5 times (in some experiments, 10 times), and the volume velocity increased by 3-10 times (in some experiments 30 times) (Fig. 3a). Incidentally, the value of the reaction to asphyxia and to other factors depends to a certain degree on the initial blood flow: with low initial values the reactions were significantly more marked than when the initial blood flow was high.

Injection of adrenalin, noradrenalin, histamine, and other biologically active substances gives rise to opposite changes of BP, but is always accompanied by definite reduction of the resistance of the vascular bed of PA and an increase in the blood flow (by 2-4 times in the case of linear, by 2-5 times in the case of volume blood flow). Usually, in response to injection of these substances, no significant changes in respiration take place (Fig. 3b, c). That is why it can be suggested that these substances have a direct action on the resistance of the vascular bed of PA, lowering it and increasing the blood flow.

The diaphragm has a multiple arterial blood supply. According to anatomical data, each half of the diaphragm is supplied with blood from the phrenic artery, the internal mammary artery, and the 8th-12th intercostal arteries [8, 14]. It has also been shown that good correlation exists between changes in the blood flow along PA in dogs and the blood supply of the diaphragm as a whole, measured with the aid of microspheres [11]. That is why it can be tentatively suggested that the principles we discovered, characterizing changes in the blood flow along PA under different conditions, reflect adequately the dynamics of the blood supply of the diaphragm as a whole.

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CHANGES IN LECTIN-INDUCED CHEMILUMINESCENCE OF NEUTROPHILIC GRANULOCYTES AFTER HELIUM–NEON LASER IRRADIATION OF BLOOD

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Neutrophilic granulocytes (NG) are involved in the pathogenesis of ischemic heart disease, most frequently through their ability to generate active forms of oxygen (AFO) [13]. The AFO, which are responsible for antimicrobial action, may also exert a direct toxic effect on healthy tissues. They also take part in arachidonic acid metabolism with the formation of biologically highly active produc leukotrienes, thromboxanes), and their subsequent inactivation [1, 4]. The most informative method of recording AFO i measurement of chemiluminescence (CHL) [10, 12]. In ischemic heart disease a sharp rise in CHL of the leukocytes is observed, and is attributed to higher AFO production in this pathology [7].

Methods of treatment of ischemic heart disease by intravenous irradiation of the blood by helium—neon laser (HNL) have recently become widely adopted [6, 8]. The concrete mechanisms of the therapeutic action of HNL, however, still remain largely unclear. The effect of HNL on production of AFO, leukotriene B_4, and thromboxane B_2 by healthy human neutrophils have not been studied at all. The present investigation was undertaken to rectify this omission.

EXPERIMENTAL METHOD

A 19-ml blood sample was taken from the cubital vein of 33 healthy fasting donors into plastic test tubes containing 2 ml of 3.8% sodium citrate. The citrated blood was divided into two 10-ml portions and passed continuously through a siliconized rubber tube with an internal diameter of 3 mm, to simulate the blood flow in a vessel. The first portion was irradiated through a light guide located in the lumen of the tube by an LG 208A HNL (wavelength 0.633 nm) for 20 min in a segment of the tube whose temperature was kept constant at 37°C. The power of the radiation at the end of the light guide was 1 mW. The second portion was subjected to the same conditions but was not irradiated, and served as the control. After the end of the procedure, the leukocyte count and leukocyte formula were determined by the usual laboratory methods. Neutrophilic granulocytes were isolated in a Ficoll—Verografin gradient [11]. After centrifugation the cells were washed twice in Hanks' buffer, pH 7.4 (without Ca^{2+} and Mg^{2+} ions). Sedimented erythrocytes were remo by hypoosmotic shock: to 1 ml of a suspension of washed cells was added 18 ml of cold distilled water, and osmolarity was restored after 20 sec by the addition of 6.25 ml of 0.6 M NaCl. The cells were washed with buffer and their number adjusted to $5.0 \cdot 10^6$ in 1 ml of suspension. The viability of the cells, in the trypan blue test, was not less than 98%. Luminol-dependent CHL was measured on an LKB Wallac 1250 chemiluminometer with integration time of 10 sec. The readings were recorded

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