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AUTONOMIC AND CEREBRO-VASCULAR ABNORMALITIES IN MILD COPD ARE WORSENED BY CHRONIC SMOKING

Short title:
Cerebral haemodynamics and COPD

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Abstract (200 words)

Question of the study

Patients with chronic obstructive pulmonary disease (COPD) may develop hypercapnia and hypoxia, two main determinants of cerebral blood flow. We tested whether cerebrovascular regulation were: altered in mild COPD, modified by manoeuvres acutely improving autonomic cardiovascular modulation, influenced by smoking habit.

Materials/Patients/Methods

In 15 eucapnic normoxic mild COPD patients (8 smokers) and 28 age-matched controls (14 smokers) we monitored mid-cerebral artery flow velocity (MCFV), end-tidal carbon dioxide (CO_2-et), oxygen saturation (SaO_2%), electrocardiogram and blood pressure at rest, during progressive hypercapnic hyperoxia, isocapnic hypoxia, slow breathing, oxygen administration. MCFV, arterial baroreflex, and dynamic MCFV- blood pressure relationship (by phase analysis), were compared.

Results

COPD- and control smokers showed higher MCFV (when corrected for CO_2), lower cerebrovascular resistance index and lower sensitivity to CO_2, with equal sensitivity to SaO_2 and phase analysis. Arterial baroreflex was depressed in all COPD. Slow breathing and oxygen administration improved baroreflex sensitivity and reduced MCFV in all COPD.

Answer

Patients with mild COPD show autonomic dysfunction. Chronic smoking induces cerebral vasodilation and impairs cerebrovascular control. All abnormalities can be partly corrected by improving the cardio- and cerebrovascular autonomic modulation, suggesting functional autonomic abnormalities present already at an early stage.

Key words

Chronic obstructive pulmonary disease
Cerebral circulation
Hypoxia
Hypercapnia
Baroreflex sensitivity
Cigarette smoking

Introduction

In normal humans and animals, cerebral blood flow is proportional to arterial carbon dioxide (CO_2) whereas it is inversely correlated with arterial oxygen tension and oxygen saturation [1,2,3]. Within limitations, cerebral blood flow is linearly correlated with blood flow velocity, as measured by trans-cranial Doppler ultrasound [1].

Because patients with chronic obstructive pulmonary disease (COPD) have a tendency to develop hypercapnia in their natural history, and also show cardiovascular and respiratory control abnormalities [4], it is likely that these combined factors affect cerebral haemodynamics. In turn, alteration in cerebral blood flow may be an important determinant of the cognitive impairment reported in these patients [5,6], and also contribute to the worsening of the autonomic and respiratory abnormalities, by impairing the perfusion of brainstem regulatory centers. However, smoking is a common habit of COPD patients, and although only few studies (and none in COPD) have reported on the chronic effect of smoking on the cerebrovascular dynamics [7,8,9], smoking seems capable of inducing long-term cerebrovascular changes [9]. Thus, it is possible that chronic smoking may be responsible or at least contribute to impair cerebrovascular regulation.

Despite this potential relevance, very few studies so far have examined the cerebrovascular control in COPD [10,11], and, in particular, comparison with similar data in healthy control subjects, to our knowledge are lacking.

The present study aims to 1) compare the cerebrovascular hemodynamics in mild COPD patients and in healthy controls to assess the presence and the origin (organic vs functional) of possible early cardio- and cerebro-vascular control abnormalities, 2) test the effect of simple respiratory manoeuvres able to improve the autonomic control of the cardiovascular system on the cerebrovascular regulation, and 3) test the possible interaction with smoking habit.

Methods

Subjects

This study was carried out in 15 subjects with mild COPD and 28 age-matched controls. The diagnosis and classification of COPD was made according to the GOLD criteria [12]. The main anthropometric data and post-bronchodilator spirometric data of the

patients and controls are presented in table 1. Eight of the COPD and 14 of the controls were active smokers (more than 10 cigarettes/day for at least 1 year); none the remaining COPD patients was active or previous smoker. The patients were characterised by mild levels of COPD, without hypercapnia or hypoxia at rest. For ethical reasons, medications were not discontinued the day of the tests. These included beta-2 agonists (10 subjects), anticholinergic agents (11 subjects), theophylline (1 subject) and cortisone (7 subjects). Smoking was discontinued during the day of the study. The protocol was approved by the local Ethics Committee and all subjects gave informed written consent to participate in the study.

Protocol

All subjects were examined in sitting position, at a comfortable room temperature and humidity. Mid-cerebral artery blood flow velocity (MCFV) was monitored by a 2 MHz transcranial Doppler probe at a depth 35-55 mm through the temporal window (Atys Medical, Soucieu en Jarrest, France) of the non-dominant side. We also recorded the electrocardiogram (by chest leads), continuous non-invasive blood pressure by the cuff method (Portapres@, Finapres Medical Systems, Amsterdam, The Netherlands), and respiratory movements, by inductive plethysmography. We continuously measured end-tidal CO_2 (CO2-et, by COSMOplus, Novametrix, Wallingford, CT, USA) and arterial oxygen saturation (SaO_2), by a pulse oxymeter (3740 Ohmeda, Englewood, CO, USA). During the entire tests the subjects were connected to a rebreathing circuit through a mouthpiece, as previously described [13,14]. In a preliminary study in 4 COPD patients we compared the arterial C O_2 pressure (38.0±1.1 mmHg, mean ± SEM), with the noninvasive end-tidal estimate (35.3±0.4mmHg). We confirmed the absence of hypercapnia in our patients and found only minimal and expected differences between invasive and noninvasive data, all in the range expected for healthy subjects.

In each subject we recorded baseline values during 4 minutes of spontaneous breathing, 2 minutes during controlled breathing at 15 breaths/min and 2 minutes during controlled breathing at 6 breaths/min. We performed the following rebreathing tests in random order, to evaluate the cerebrovascular sensititvity to hypercapnia and hypoxia, respectively:

1. Progressive normocapnic hypoxia (SaO_2 from baseline to 80%, CO_2-et was maintained at a standard level of 38 mmHg)

2. Progressive hyperoxic hypercapnia (CO_2-et raised from baseline by +15 mmHg, SaO_2 was > 98 %).

When the sensitivity to hypercapnia was tested, oxygen was supplied to the rebreathing bag at very low flow, in order to maintain SaO_2 above 98%. During the first 2 minutes of this procedure the carbon dioxide was maintained constant, and the data obtained were compared with baseline data (while breathing room air) to test the effect of steady-state oxygen administration. During progressive hypoxia, the CO_2 levels were clamped by passing a variable part of the expired air into a reservoir filled with soda lime, under continuous visual control of CO_2-et. During all these recordings we continuously recorded all cardiovascular and cerebrovascular signals.

Data acquisition and analysis

All signals were continuously acquired on a personal computer (Macintosh Powerbook, Apple, Coupertino, USA) at the frequency of 600 samples/channel. The cerebrovascular sensitivity to hypoxia or hypercapnia was obtained from the slopes of the linear regression of mean MCFV vs SaO_2 or CO_2-et, respectively, for each breath. The arterial baroreflex sensitivity was calculated from the time series of RR interval and systolic blood pressure, obtained at baseline and during controlled breathing at 15 and 6 breaths/min, by the so-called "alpha index" (by autoregressive spectral analysis of RR interval and systolic blood pressure) [13,15]. The MCFV was evaluated during each sequence, and the values were corrected for the corresponding CO_2-et (MCFV/CO_2-et) [15,16]. We also corrected the MCFV for the contribution of mean blood pressure (MBP/ MCFV), thus obtaining an index of cerebrovascular resistance [17], and finally we considered the combined effects of blood pressure and CO_2-et on MCFV (MBP / MCFV/CO_2-et) [17].

To test the dynamic relationships between blood pressure and MCFV, we applied the transfer function phase analysis. The measurement of the phase delay between MCFV and mean blood pressure [18] has been proposed as a method to test dynamic cerebrovascular regulation, under undisturbed conditions. According to the high-pass filter model of cerebral regulation, variations in MBP should be transmitted to MCFV [19,20]. Accordingly, we expected that MCFV was leading MBP under normal conditions. In the case of disturbed regulation, the phase shift between both parameters would have approached 0, ie, MCFV was following MBP changes passively [19,20]. We used an

autoregressive spectral algorithm [19] to calculate the amplitudes and phases (expressed in degrees) of the recorded parameters.

Statistical analysis

Data are presented as means±SEM. Differences between the COPD and controls, and between smokers and non smokers were assessed by Analysis of variance (factorial design on two factors, to test for the effects of COPD and smoke) [21]. Differences between different breathing rates in different groups were assessed by a 2-way mixed-design (repeated measures for tests within groups and factorial between groups) analysis of variance [21]. P= 0.05 was the limit for statistical significance.

Results

Baseline data (figure 1)

At baseline, we found significant ($p<0.025$ or greater) overall differences on analysis of variance between the 4 different groups of subjects. The differences were essentially due to smoking, which induced a marked resting cerebral vasodilation, evidenced by a significant ($p<0.01$) increase in resting MCFV, which remained significant ($p<0.005$) even after correction for CO_2-et levels or also when corrected for mean blood pressure and thus expressed in terms of index of cerebrovascular resistance ($p<0.05$), and finally, also when this index was corrected for the effect of CO_2-et levels ($p<0.025$). Conversely, no significant differences were seen when considering the presence of COPD.

Sensitivity of MCFV to hypoxia and hypercapnia (figure 2)

Progressive isocapnic hypoxia and progressive hyperoxic hypercapnia increased MCFV, in both COPD and controls. The average slope of the curves showed no significant differences in the two groups. Conversely, significant differences were observed by effect of smoking: the smoker subgroups showed both significant ($p<0.05$) reduction in CO_2 sensitivity. Furthermore, during progressive hypercapnia we found in 5/8 COPD-smokers and in 5/14 control-smokers a plateau at higher levels of CO_2-et, indicating a progressive lower sensitivity for higher levels of CO_2-et. An example is shown in figure 3. The occurrence of this plateau was also associated with higher resting values of MCFV.

Effect of respiratory manoeuvres (figure 4)

As compared to spontaneous breathing, controlling the breathing rate at 15 breaths/min induced a parallel reduction in both the MCFV and CO_2-et in both COPD and control subjects, thus leaving unchanged the MCFV when corrected for CO_2-et. However, when the subjects breathed at 6 breaths/min, the drop in MCFV in the COPD group (and to a lower extent in control subjects), remained significant even after correction for CO_2-et. At baseline, baroreflex sensitivity was reduced in all COPD patients (5.7±0.8 vs 10.1±1.0 ms/mmHg, p <0.01), without differences between smokers and non smokers (6.4±1.1 vs 5.2±1.1, respectively, p:ns). Controlled breathing at 6 breaths/min significantly increased baroreflex sensitivity in all subjects (8.9±1.7 ms/mmHg in COPD, p<0.05 vs spontaneous breathing and p: ns vs control subjects, and 14.3±2.0 ms/mmHg in control subjects, p <0.01 vs baseline), whereas controlled breathing at 15 breaths/min did not induce significant changes vs spontaneous breathing. Smokers and non-smokers showed identical trends with respiratory manoeuvres.

Effect of oxygen administration

Although resting oxygen saturation levels were normal in both groups, oxygen administration increased SaO_2 in both groups and significantly reduced MCFV/CO_2-et values in the COPD group, while no changes were seen in controls (figure 5). Smokers and non-smokers showed identical trends.

Phase analysis between blood pressure and MCFV

In the Low-frequency range, MCFV was leading MBP in both groups, with similar phase delays (65.3±8.6° vs 53.4±6.6°, respectively, p: n.s., figure 6). Controlled breathing at 15 breaths/min and at 6 breaths/min, did not induce significant changes nor differences between the two groups. Smokers and non-smokers showed identical values.

Discussion

Main findings

This study shows for the first time, to our knowledge, that patients with even mild levels of COPD have already evident alterations in cerebrovascular control, characterised by a higher resting MCFV levels and a lower index of cerebrovascular resistances, even after correction for CO_2. These abnormalities were limited to the subgroup of COPD with smoking history, suggesting that the smoking habit could have been responsible for most

of the changes observed in the cerebrovascular modulation. However, baseline baroreflex sensitivity was equally reduced in smokers and non-smokers, suggesting that autonomic dysfunction was to some extent independent of smoking habit. The sensitivity to CO_2 was also reduced in the smoking subgroup of COPD at higher CO_2 levels. We speculate that these abnormalities might have a potential compensatory role, preventing excessive vasodilation in the presence of hypercapnia. The improvement of these abnormalities by simple respiratory manoeuvres and by oxygen administration, indicates that these abnormalities are functional and could be reversed by appropriate therapy.

Increased resting MCFV in mild COPD patients

Subjects with even mild signs of COPD, seem to have a substantial increase in MCFV, suggesting, within the limitations intrinsic in the trans-cranial Doppler measurements [3], an increased cerebral blood flow. This finding is clearly due to the smoking habit, common in COPD, as it was also present in smokers with normal spirometry. The increase was more evident when MCFV data were corrected for the actual value of CO_2-et. This is an essential correction to be done, as it is well known that CO_2 is the major determinant of MCFV [2,3,15,16], and even a mild hyperventilation may actually induce an artifactual reduction in MCFV. For practical reasons, CO_2 had to be estimated from end-tidal values. In theory, due to ventilation/perfusion abnormalities typical of advanced COPD, CO_2 levels could have been underestimated, leading to artifactual increases in MCFV/CO_2 ratio. While this possibility should be taken into account (particularly in more compromised patients), the subjects of the present study were all normocapnic (or even frankly hypocapnic), and only minor and expected differences between intra-arterial and end-tidal values were found in our preliminary testing. Furthermore, all our subjects had only minor pulmonary dysfunction, indicating that our estimation of pCO_2 by end-tidal values was not substantially biased. Additionally, the other finding of a reduced MBP/ MCFV ratio, considered as an index of cerebrovascular resistance resulted lower in COPD-smokers even without correction for CO_2.

An increase in MCFV may result from an altered sensitivity to hypoxia or hypercapnia. Our results confirm that the subjects showing an increase in resting MCFV (essentially the smoker groups) had also a reduced sensitivity to CO_2. In 5/8 of these COPD patients and in 5/14 control smokers we found that higher resting values were associated with a plateau in the relationship between MCFV and CO_2-et (figure 3). This suggests a lower ability to vasodilate the brain vessels for higher levels of CO_2-et. A recent

study also showed that a logistic function would better fit the MCFV – CO_2 relationship, indicating a general tendency, even in healthy subjects, to reduce the sensitivity for higher levels of CO_2-et [22]. Therefore, our finding is probably an amplification of a normal phenomenon, whose finalism could be the prevention of extreme vasodilation at higher levels of CO_2 and/or MCFV. It is well known that extreme cerebral vasodilation, due to hypercapnia or hypoxia (typically occurring at high altitude) can lead to severe headache and even predispose to cerebral oedema [23,24]. This may be relevant in COPD, as hypercapnia (and often hypoxia) is a common complication in this disease. On the other hand, these findings indicate that estimates of sensitivity to CO_2 may vary depending on the range of data used for the calculation. In our case (being none of the COPD patients hypercapnic), we could examine the cerebrovascular pattern of response to CO_2 from low levels till frank hypercapnia, thus avoiding all possible bias.

The cerebrovascular changes we have seen were essentially confined to the sub-groups of smokers, with or without COPD. While several studies [7,8,9], evaluated the acute effect of smoking on cerebrovascular dynamics, only very few reported baseline comparisons between smoker and non-smoker subjects, no previous studies of this type exist in COPD to our best knowledge, and in none of the studies were MCFV values corrected for CO_2, ventilation or blood pressure. To the limited extent that data could be compared, our present results are in agreement with previous data reporting an increase in resting MCFV values in smokers at baseline [9].

Autonomic disturbances in COPD patients

Autonomic abnormalities have been consistently found in COPD. These span from a reduction in heart rate variability [25,26,27] a reduction in respiratory sinus arrhythmia [27] and a reduction in baroreflex sensitivity [28,29], together with a direct increase in muscle sympathetic nerve activity [30,31]. In the present study we found that our COPD patients had depressed baroreflex sensitivity, even regardless the history of smoking, and regardless of the drugs assumed. While the acute effect smoking is a clear reduction in baroreflex sensitivity [32], the chronic effect is much less evident [33]. Similarly, from our data it did not result as a significant determinant of the observed reduction in baroreflex sensitivity in our COPD. In healthy subjects and patients with autonomic dysfunction we previously found that reducing the breathing rate can acutely improve baroreflex sensitivity and improve autonomic function [13,34,35]. In the present study, the controlled breathing at slower rate (6 breaths/min) induced an evident and significant reduction in MCFV in

COPD patients, that became similar to that of controls. This was associated with an increase in baroreflex sensitivity toward normal levels, suggesting that the changes observed occurred together with an improvement in the autonomic cardiovascular modulation. These findings were not due to a change in CO_2-et induced by the change in minute ventilation, as the MCFV changes remained evident after correcting MCFV for CO2-et levels. The reduction in MCFV induced by slower breathing indicates that the autonomic abnormalities were likely functional and not the consequence of an established neuropathy.

During spontaneous breathing the phase angle between the oscillations in MCFV and mean blood pressure, in the 0.1Hz range, was similar in COPD and controls, and no changes were seen with controlled breathing at different rates. The interpretation of this phase angle is controversial: while some authors consider it as a marker of dynamic auto-regulation [18], we have previously suggested that it may instead depend upon the autonomic control of the cerebral vessels [19,36]. A larger phase angle implicates the intervention of some modulating factor (either intrinsic to the vessels or with the implication of the autonomic nervous system), whereas a smaller angle indicates a more passive transmission between main arteries and blood vessels. In fact, a smaller angle is typically seen in severe autonomic neuropathy [19]. In this study, we found no reduction in phase angle in subjects with COPD, despite depressed baroreflex sensitivity, confirming our other finding that the control of cerebrovascular circulation was not markedly affected in our COPD. Similarly, COPD subjects reduced their resting MCFV while breathing oxygen (figure 5), indicating that even in the normoxic range, there was an effect of oxygen at cerebrovascular level.

Thus, even patients with mild COPD have autonomic dysfunction, that could predispose to abnormalities in the cerebrovascular regulation. In turn, this could potentially amplify the smoke-dependent abnormalities in cerebrovascular dynamics. While a frank neuropathy has been described in severe COPD [37], our findings of an immediate modification with breathing manoeuvres suggest that these abnormalities resulted from a still functional (and reversible) disturbance in patients with limited clinical involvement. The clinical implications of an alteration in the autonomic nervous system and cerebrovascular dynamics are clear when considering the recently reported high sensitivity of the solitary tract nucleus to changes in blood flow [38]. Additionally, the alteration in cerebrovascular dynamics may affect cognitive performance [5,6,39]. This, together with

blood gas abnormalities occurring in the natural history of COPD, may be an important factor conditioning cognitive impairment seen in these patients [39].

Limitations of the study

In the present study we reported cerebrovascular and autonomic abnormalities in mild COPD. Whether this is the beginning of a progression cannot be established by our data, and further studies are needed in a more compromised population to assess whether and to what extent these abnormalities further progress, though clinical and technical difficulties involved in correct CO_2 assessment makes it more difficult in more compromised patients. While an invasive estimation of arterial pCO_2 could have provided a more reliable measurement, this was performed only in a small subgroup of patients at baseline, being impossible to follow the data over time (eg during rebreathing). Although in theory our findings could have been influenced by a ventilation/perfusion mismatch typical of advanced COPD, the comparison with invasive data showed normal CO_2 values ruling out this possibility, at least for the group of patients we have examined. This methodological problem should be taken in due account in more compromised patients. The present results suggest also a possible role of CO in the modifications observed, that deserve further clarification. Although the changes observed in cerebrovascular dynamics appear a specific effect of smoking, their relationship with the further development of autonomic abnormalities in COPD need to be further studied, however, due to the importance of smoking in the aetiology and progression of COPD it appears that these findings underline further the importance of smoke abolition in these patients.

Conclusions

We provide evidence that patients with even mild levels of COPD present an initial degree of autonomic dysfunction, which in COPD-smoker is associated with impaired cerebrovascular regulation. Functional cerebrovascular abnormalities appear to be typical of smokers, even regardless of COPD. The immediate improvement of these indices with simple manoeuvres able to produce a transient improvement in the autonomic modulation of the cardiovascular system suggests that the abnormalities we have described are to a great extent functional and could be probably reversed with appropriate therapy. It is likely that our data have clinical relevance, as an increase in resting MCFV, together with a reduced autonomic control may be one of the factors predisposing these patients a higher cerebrovascular risk [4,39]. The other novel finding of the presence of cerebrovascular

abnormalities in the COPD-smokers should provide an additional argument in favor of smoke abolition. Further studies are needed to establish the time course of these abnormalities in more severe patients.

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Table 1.

COPD patients and controls at baseline

		COPD			Healthy Controls		
		smokers	non-smokers	all	smokers	non-smokers	all
N		8	7	15	14	14	28
Sex	(m/f)	5/3	5/2	10/5	8/6	5/9	13/15
Age	(yr)	50.6±2.3	54.0±5.1	52.2 ± 2.6	47.6±1.8	46.7±2.9	47.2±1.7
Size	(cm)	177.2±1.7	172.8±3.5	175.2 ± 1.9	170.6±2.1	174.7±1.7	172.6±1.4
Weight	(kg)	79.2±3.3	72.0±3.4	75.8 ± 2.5	72.2±4.8	70.4±2.8	71.3±2.8
BMI	(kg/m^2)	25.3±1.3	24.0±0.5	24.7±2.77	24.7±1.5	22.9±0.7	23.9±0.8
FEV1 % predicted		75.6±3.9	75.5±3.0	75.6 ± 2.1**	82.5±2.6	83.1±2.2	82.8±1.7
FEV1/FVC		67.7±0.8*	66.1±1.8	67.0 ± 0.9***	89.4±2.1	85.8±2.7	87.6±1.7
Smoke (pack-year)		20.1±2.7	--	--	16.0±2.1	--	--

*: p<0.05, **: p<0.01, ***: p<0.001 vs healthy controls

FIGURE LEGENDS

Figure 1

Resting levels of MCFV, corrected for the values of CO_2-et and of mean blood pressure, in COPD and healthy controls (sm+: smokers, sm-: nonsmokers).

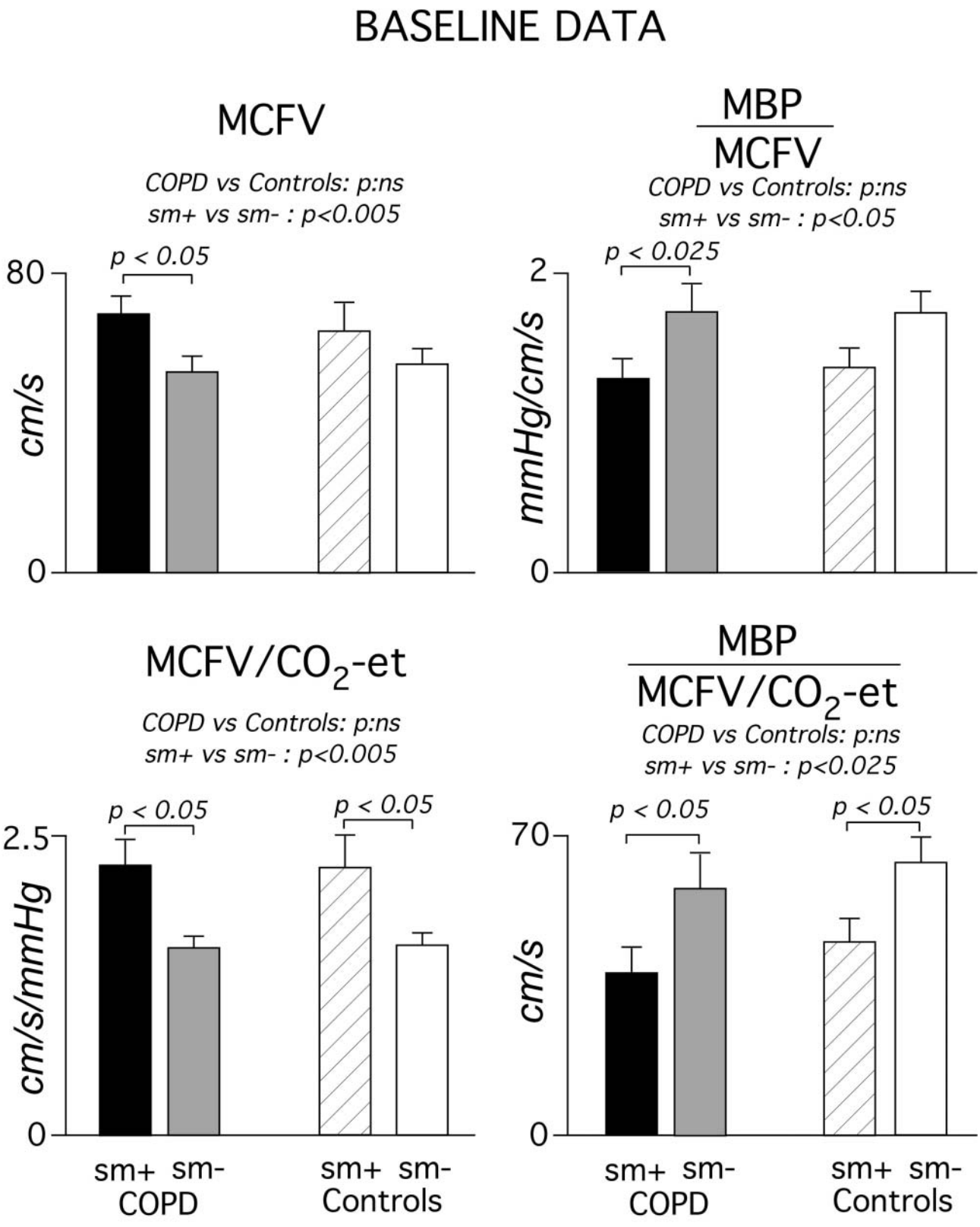


Figure 2

Sensitivity to CO_2 and to Oxygen, in COPD and healthy controls (sm+: smokers, sm-: nonsmokers).

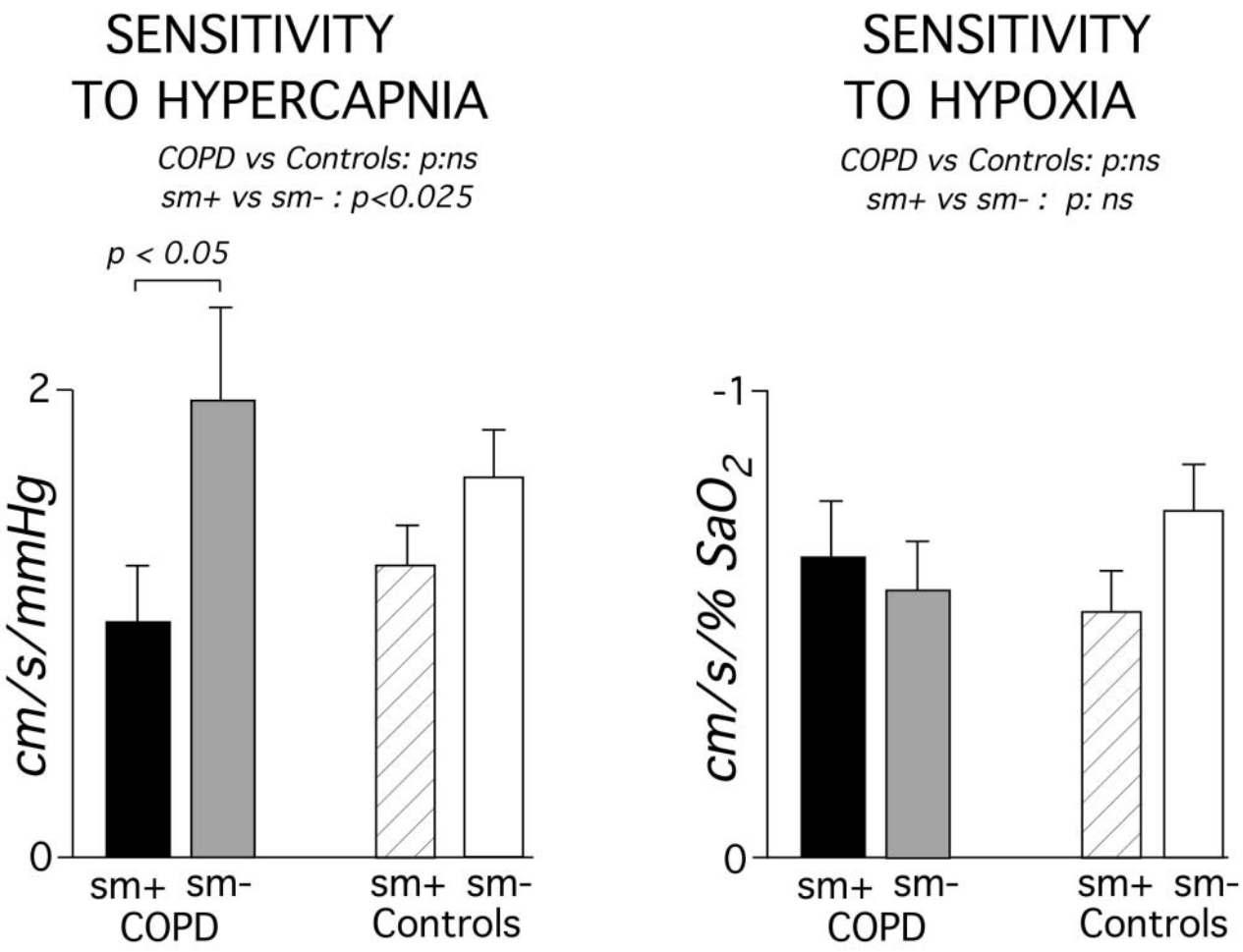


Figure 3

Example of the raw data obtained in one COPD and in one control subject during progressive hypercapnia. Notice the higher starting level of MCFV despite similar starting levels of CO_2-et, and the plateau for increasing levels of CO_2-et in the COPD subject.

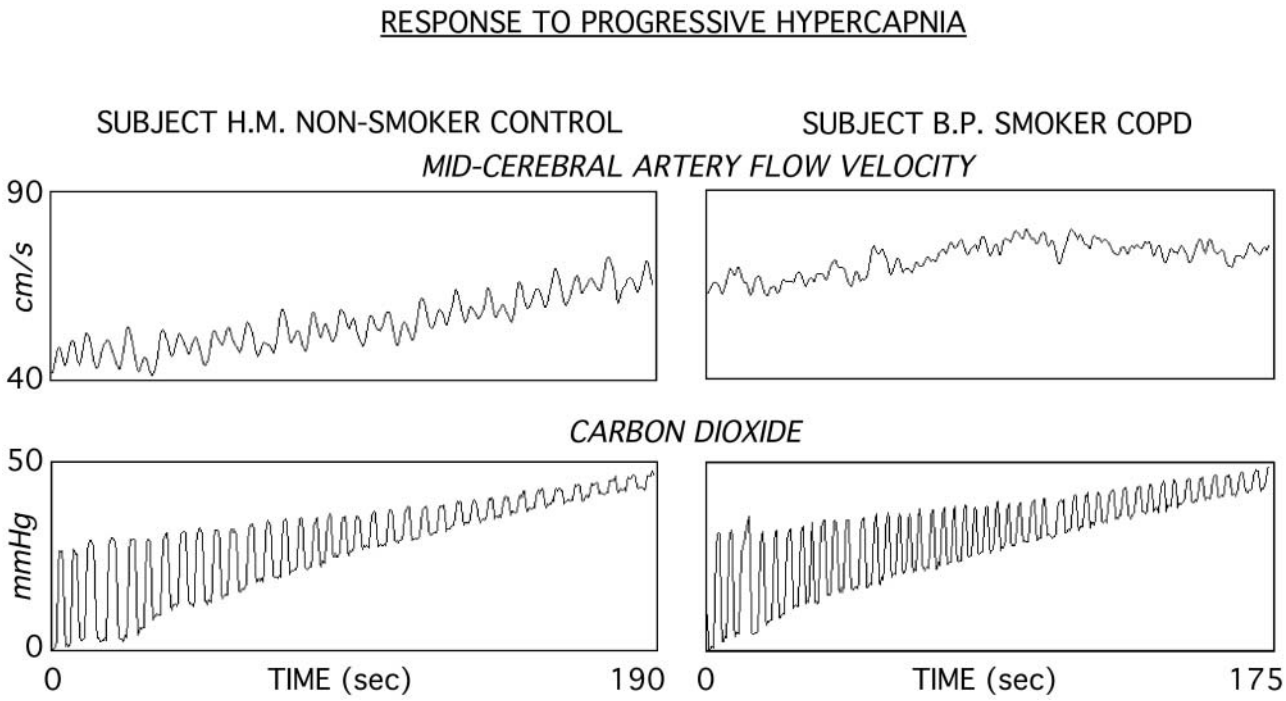


Figure 4

Effect of different breathing rates on raw MCFV and after correction for CO_2-et. Notice that, after correction for the levels of CO2-et, MCFV was decreased only during slow breathing, thus indicating that a modification in the cerebrovascular regulation occurred at this rate independently from CO_2-et.

EFFECT OF CONTROLLED/SLOW BREATHING

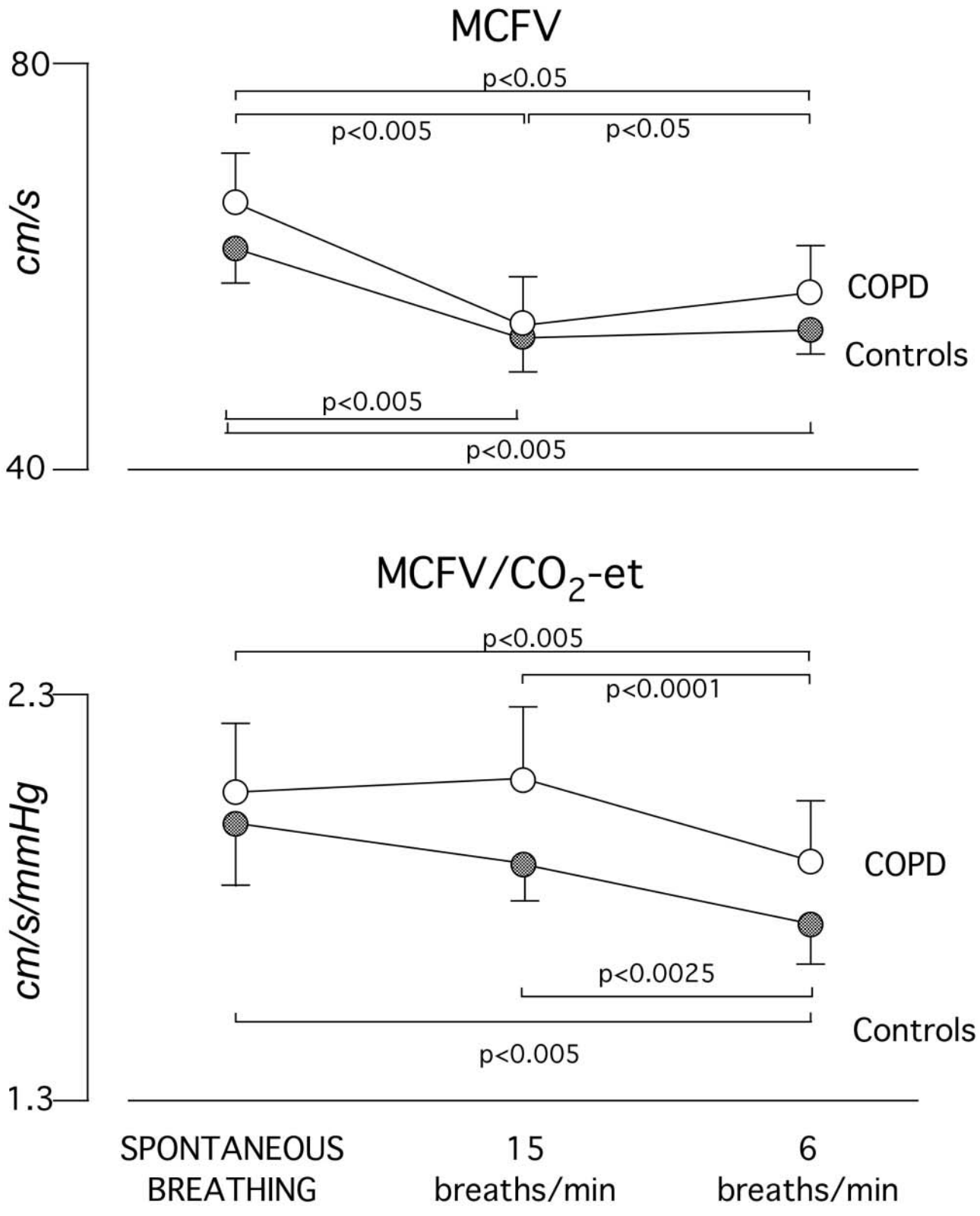
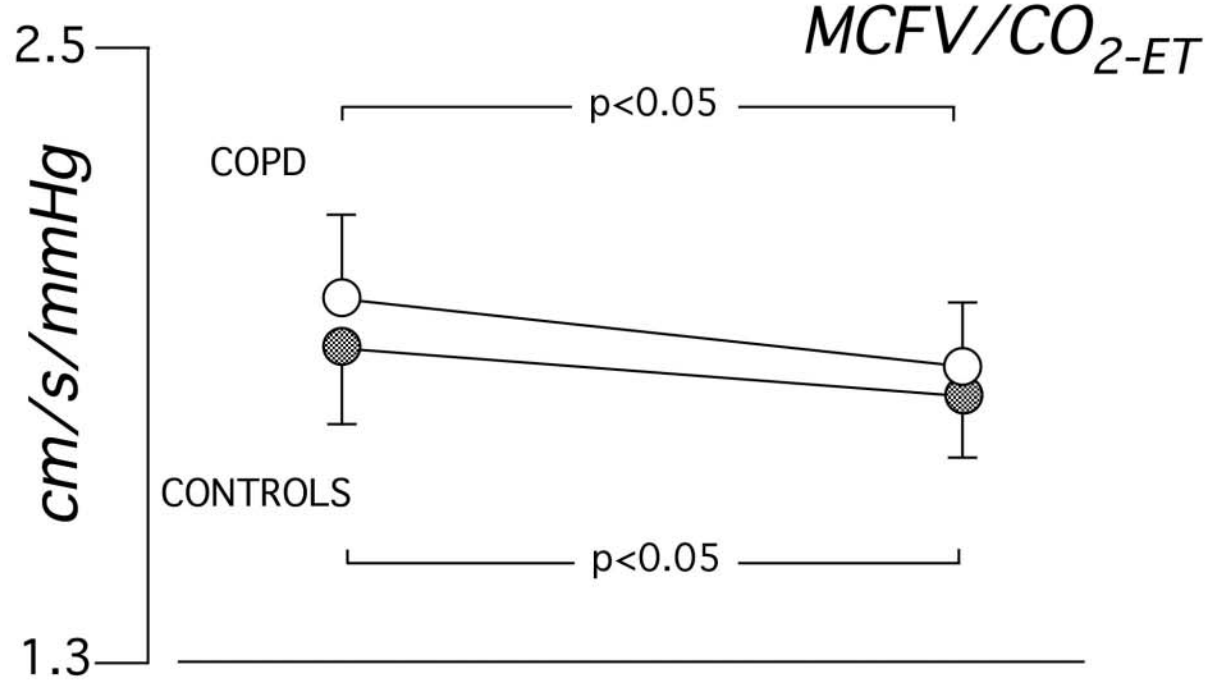


Figure 5

Effect of oxygen administration on MCFV data, corrected for CO_2-et. Despite all subjects were normoxic, and thus hyperoxia induced only a minor increase in SaO_2, this intervention reduced MCFV in the COPD and control groups.

EFFECT OF OXYGEN ADMINISTRATION



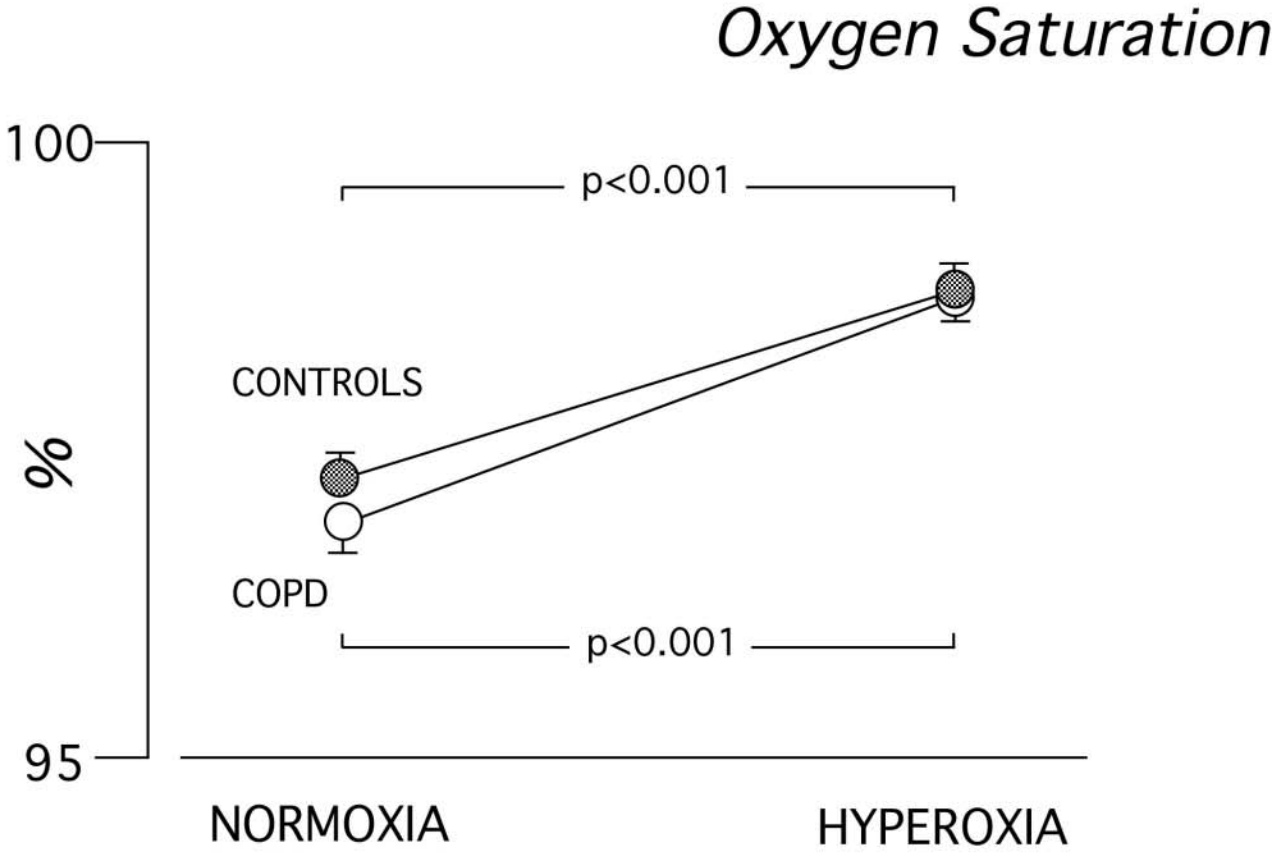


Figure 6

Results of phase analysis. The circle indicates the period of the oscillation (0.1Hz, equivalent to 10 seconds). The arrow indicate the order of precedence of oscillations. The position of the black circles indicate the relative phase delay of the oscillations in MCFV in relation to those in blood pressure. The thick lines on the circumferences indicate ± SEM from the average position. The 0.1Hz oscillations in MCFV always preceded those in mean blood pressure, by a similar extent in COPD and controls (see also the text and [19] for a further explanation of this methodology).