Exhaled nitric oxide and exercise tolerance in severe COPD patients

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Abstract
To answer the question as to whether pulmonary rehabilitation programs (PRP) induced increase in exercise tolerance (ET) is associated with increased levels of exhaled nitric oxide (eNO) in COPD patients of different degrees of severity, we designed a prospective and controlled study. Forty-seven stable COPD patients underwent an 8-week outpatient multidisciplinary PRP including supervised incremental exercise. Fractional eNO concentration (FENO) and peak work-rate (W_peak) were assessed baseline (T1), at the end of 1-month run-in period (T0), and after (T1) the PRP. Lung function, walking test, health-related quality of life (HRQL) were also recorded. Patients were divided into three groups according to disease severity: 17 severe [FEV1 35 (5)% pred] COPD patients, seven of them with cor pulmonale: 15 mild [FEV1 78 (6)% pred], and 15 moderate [FEV1 56 (6)% pred] COPD patients. FENO did not differ at T1 and T0 (mean absolute change (SD): 0.03 (0.09) 95% CI -0.01, 0.16, 0.06 (1.03) 95% CI 0.03, 0.75 and 0.05 (0.06) 95% CI 0.02, 0.11 ppb in mild, moderate and severe patients, respectively). As compared to T0, both W_peak (by 17, 15 and 10%, respectively) and FENO (by 29, 24 and 16%, respectively) significantly increased in all groups, but not in patients with cor pulmonale. A significant correlation between pre- and post-PRP changes in W_peak and FENO was found both in mild to moderate (r = 0.79, P < 0.00001) and severe (r = 0.76, P < 0.001) COPD patients. After a PRP, improvement in ET is associated with an increase in eNO also in most severe COPD patients, but not in those with cor pulmonale.

Keywords
Chemiluminescence analysis; exercise training; airway obstruction.

INTRODUCTION
Exhaled nitric oxide (eNO) has been proposed as a non-specific bio-marker in animals and humans (1). Low eNO levels have been reported in severe chronic obstructive pulmonary disease (COPD) (2), especially in patients with cor pulmonale (3).

In healthy subjects, eNO has been shown to be a marker of physical fitness (4). More recently, pulmonary rehabilitation program (PRP) related increase in exercise tolerance (ET) was found to be associated with an increase in eNO in mild-to-moderate COPD patients (5).

Benefits of PRP have been reported in COPD patients of different severity (6). We wondered whether changes in eNO associated with increases in ET might also be observed in most severe COPD patients. Indeed, the recent development of new therapeutical approaches such as lung transplantation and lung volume reduction surgery make severe COPD patients, even those with chronic respiratory failure, candidates for rehabilitation programs (7).

METHODS
The study was approved by the Ethical Committee of Salvatore Maugeri Foundation IRCCS and patients gave their informed consent to participate in the study.

Patients
Forty-seven consecutive, male, stable COPD patients (age 67 ± 7 years) referred to a multidisciplinary outpatient PRP at the Pulmonary Rehabilitation unit of S. Maugeri Foundation, Scientific Institute of Gussago, were enrolled. They were divided into three groups according to disease severity as defined by the European Respira-
NITRIC OXIDE IN EXERCISED COPD PATIENTS

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Society (ERS) guidelines (8): mild [n=15, FEV1 78 (6)% pred], moderate [n=15, FEV1 56 (6)% pred] and severe [n=17, FEV1 35 (5)% pred]. All patients were ex-smokers and none of them had any history of atopy. At the time of the study, all of them were clinically stable and were receiving their regular treatment with inhaled anticholinergics and rescue short-term-β2-agonists but neither systemic nor inhaled steroids. Seven out of 17 patients in the severe group were on long-term domiciliary oxygen therapy and suffered from cor pulmonale as assessed by echocardiography.

Assessment

The study was prospective and controlled, the patients serving as controls of themselves. Measurements of eNO and cycloergometry were performed at T-1, at the end of a 1-month run-in period (T0), and after the completion of PRP (T1). To further assess the clinical effects of PRP, the 6-min walking distance test (6MWD) (9) and health-related quality of life by means of the St. George Respiratory Questionnaire (SGRQ) (10) were measured at T0 and T1. Data were collected by personnel not involved in the study. Furthermore, physicians evaluating exercise tests were blind to eNO measurements.

Lung function: Static and dynamic lung volumes were measured by means of a constant volume body plethysmograph (Medical Graphic Corp. St. Paul, MN, U.S.A.) with the patient in sitting position according to standard procedure (11). Exercise test: Symptom-limited incremental exercise test was performed under EKG monitoring, on an electrically braked cycloergometer (Ergometrics 800S, Sensormedics, Yorba Linda, CA, U.S.A.) using the standard 1-min incremental cycle exercise protocol (10 W each minute) as previously described (5). Mixed-expired gas data, minute ventilation V̇E, and breathing pattern, were continuously monitored. At rest and at 1-min intervals during exercise, patients were asked to point to a number or phrase on a 10-point Borg scale set in large type on a sheet in front of them (12).

Exhaled NO: Resting exhaled NO was assessed on-line by means of a chemiluminescence analyzer (LR 2000 series, Logan Research, Rochester, U.K.) as recommended and previously described (2,3,5,13). The mean value of three reproducible (less than 5% within patient variability) readings of eNO plateau (FENO) was considered. Pulmonary Rehabilitation Program

The PRP was a multidisciplinary program consisting of three 3-h sessions per week for 8–10 weeks, including supervised incremental exercise, abdominal, upper and lower limb muscle activities, shoulder and full-arm circling, patient’s specific education which has been described elsewhere (5,14).

Statistical analysis

All data are shown as mean and standard deviation (SD) or standard error (SE) as specified. One-way ANOVA for repeated measures was used to test within patients time course of results: post hoc test was then applied when appropriate. Wilcoxon matched paired test was added to test differences for non-parametric measures. Spearman analysis was then used to evaluate the linear correlation coefficient between FE(NO) and other functional parameters. A P value of less than 0.05 was considered to be statistically significant.

RESULTS

Characteristics of patients in the study and results are shown in Table I. As expected, baseline ET was slightly inversely correlated with airway obstruction (FEV1 vs Wpeak: r=-0.37, P=0.009). No differences in ET was found between T-1 and T0. After PRP, Wpeak significantly increased by 17, 15 and 10% in mild, moderate and severe patients, respectively, whereas the increase in 6MWD was not significant. The lowest increase in Wpeak observed in severe patients was mostly due to seven patients with cor pulmonale who did not show any change in ET [from 61 (19) to 62 (24) W at T0 and T1, respectively, mean change (SD): -1.4 (6.9) 95% CI -1.9 8.1 W], the other 10 severe COPD patients without cor pulmonale, showing a mean 14% increase in Wpeak.

Compared with T-1 and T0, at T1 D and F at the same workload (isowatts) were significantly reduced in all groups. SGRQ (total score) significantly improved by 19, 15 and 10% in mild, moderate and severe patients, respectively (Table I).

The mean values of FE(NO) measured over time are also shown in Table I. Resting FE(NO) assessed at T-1 and T0 were very similar (mean absolute change (SD): 0.03 (0.9) 95% CI 0.01, 0.16, 0.06 (1.03) 95% CI 0.03, 0.75 and 0.05 (0.06) 95% CI 0.02 0.11 ppb in mild, moderate and severe patients, respectively). Baseline FE(NO) was higher in mild and moderate than in severe patients. At T1 FE(NO) significantly increased by 29, 24 and 16% in mild, moderate and severe COPD, respectively (Table I). The lowest increase in FE(NO) observed in severe patients was due to seven COPD patients with cor pulmonale [from 5.2 (0.9) to 5.7 (1.4) ppb at T0 and T1, respectively, mean change (SD): -0.5 (0.8) 95% CI -0.06 0.91 ppb]. The other 10 severe...
patients without cor pulmonale showed a post-PRP mean increase in FENO by 22%.

There was significant correlation between pre- and post-PRP changes in Wpeak and in FENO in the whole group (r = 0.78, P < 0.0001; Fig. 1—panel A) as well as in the mild to moderate (r = 0.79, P < 0.00001; Fig. 1—panel B) and severe (r = 0.76, P < 0.001; Fig. 1—panel C) patients. No significant relationship was observed between changes in FENO and changes in 6MWD or SGRQ in any group of patients.

DISCUSSION

This study shows that improvement in ET following an outpatient PRP is associated to a parallel increase in resting eNO in COPD patients of different degrees of severity. This increase is also observed in COPD patients with most severe airway obstruction, but not in those with cor pulmonale.

This study confirms that PRP may induce benefits in ET of COPD patients independent of the degree of severity (6). Nevertheless, severe patients with cor pulmonale in our study did not show such positive results.

In a previous study (5), after a multidisciplinary PRP, mild-to-moderate COPD outpatients showed an increase in eNO correlated with the improvement in ET, thus suggesting FENO as a marker of physical fitness (15). This study extends those results (5) to severe COPD patients without cor pulmonale. The highly significant correlation between pre- and post-PRP changes in Wpeak and in FENO was independent of airway obstruction (see Fig. I). An indirect evidence of the relationship between ET and FENO is also given by the fact that patients with cor pulmonale neither gained any benefit from PRP, nor showed any increase in eNO (Fig. I, open triangles).

Different from Wpeak, no significant relationship was observed between post-PRP changes in FENO and 6MWD. This reflects differences in the modalities of exercise tests, the 6MWD being more related to the endurance capacity than the incremental test. The lack of relationship between changes in eNO and in HRQL is not surprising, probably reflecting the comprehensive

| Table I. Demographic, anthropometric, physiological characteristics of patients and effects of PRP. Data are given as mean and (se) |
|---------------------------------|---------------------------------|---------------------------------|
|                                | Mild (n=15)                      | Moderate (n=15)                 | Severe (n=17)                  |
|                                | T-1 | T0  | T1  | T-1 | T0  | T1  | T-1 | T0  | T1  |
| Age (years)                    | 69  | 67  | 66  | 28  | 27  | 26  | 28  | 27  | 26  |
| BMI                            | 28  | 27  | 26  | 28  | 27  | 26  | 28  | 27  | 26  |
| FEV1 (%)                       | 77  | 57  | 34  | 77  | 57  | 34  | 77  | 57  | 34  |
| % predicted (%)                | (6) | (6) | (7) | (6) | (6) | (7) | (6) | (6) | (7) |
| Wpeak (W)                      | 95  | 91  | 107†| 83  | 82  | 94†| 65  | 68  | 75†|
| (score)                        | (18)| (23)| 29  | (22)| (19)| 26  | (22)| (28)| (36)|
| Fiso-watt (score)              | 7.3 | 77  | 6.0  | 7.6 | 6.4 | 5.5  | 8.3 | 8.0 | 6.1  |
| (m)                            | (1.1)| (1.9)| (0.5)| (2.0)| (2.2)| (0.7)| (0.8)| (1.3)| (0.6)|
| 6MWD (m)                       | 463 | 502 | 473 | 503 | 473 | 503 | 380 | 424 |
| SGRQ (total)                   | 38  | 31  | 39  | 33  | 39  | 33  | 48  | 43  |
| FENO (ppb)                     | 7.3 | 77  | 100†| 5.3 | 6.2 | 77†| 5.5  | 5.7 | 6.6†|
| (A) Anova P < 0.05 between groups. |
| (B) Post hoc test P < 0.005 (vs T0 and T-1) |
| (C) Post hoc test P < 0.01 (vs T0 and T-1) |
| (D) Post hoc test P < 0.05 (vs T0 and T-1) |

Wpeak = peak workload, 6MWD = 6 min walking distance walk, SGRQ = St. George Respiratory Questionnaire, D = iso-load perceived dyspnoea by BORG scale, F = iso-load perceived leg-fatigue by BORG scale.
nature of the PRP and the fact that HRQL depends on more than just exercise ability (14).

In conclusion, this study answered the initial question in that post-PRP improvement in ET is associated to changes in eNO in most severe COPD patients, but not in those with cor pulmonale. What is the relative role of eNO and physical conditioning in these patients must be elucidated by further studies.

REFERENCES

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