
Evaluation of Fractional Exhaled Nitric Oxide During SARS-CoV-2 Infection

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Fractional exhaled nitric oxide (FeNO) is a noninvasive marker of type 2 inflammation. It is increasingly measured in the diagnosis and follow-up of allergic diseases such as asthma. High levels of FeNO are correlated with asthma attacks and are being evaluated in other respiratory diseases. Respiratory infections also have a significant effect on FeNO through an as yet unknown mechanism [1-3]. The increase or decrease in FeNO during viral infection is unclear, although it seems to be virus-dependent [4]. The unknown and variable clinical progress of SARS-CoV-2 infection from asymptomatic to death has forced us to investigate new biomarkers that could predict the natural course of the disease.

Aiming to assess the potential role of FeNO as a marker of severity in cases of COVID-19, we included consecutive patients over 18 years of age who received care in the emergency department (ED) at Fundación Jiménez Díaz Hospital, Madrid, Spain for SARS-CoV-2 infection from January to June 2021. All patients had a positive reverse transcriptase-polymerase chain reaction (RT-PCR) result and/or positive antigen test result. We included a control group consisting of patients with respiratory symptoms and a negative RT-PCR result for SARS-CoV-2. The local ethics committee approved this study. All patients provided their written informed consent to be included.

FeNO was measured in duplicate using the Evernoa device (Eversens) [5]. The first measurement was taken at baseline, before any therapeutic intervention in the ED; the second was performed at least 10 days later (recovery from infection). The data collected included demographic, clinical, and disease-related characteristics, presence of atopy (patient-reported), as well as comorbidities (eg, pneumonia), treatment, hospital and intensive care unit (ICU) admission if required, blood analysis, and chest x-ray findings. If the patient was admitted, FeNO was measured every 48-72 hours until discharge. The control group included 18 patients.

Quantitative variables were expressed as mean (SD), and qualitative variables as absolute and relative frequencies.

Intergroup comparisons were performed using the χ^2 test or Fisher exact test for qualitative variables and ANOVA or Kruskal-Wallis test for quantitative variables. *P* values <.05 were considered significant

Eighty-two patients were included and divided into 3 groups according to the severity classification of the WHO [6], as follows: 25 patients with mild infection (uncomplicated upper respiratory tract viral infection), 26 with moderate pneumonia (no initial need for supplemental oxygen), and 31 with severe pneumonia ($\text{SpO}_2 \leq 93\%$ in room air at baseline). Patient characteristics and laboratory findings are summarized in the Table and Figure S1 in the supplementary material.

Overall, mean FeNO for the study population was in the normal range during infection, with no significant variation during the recovery phase (Table). There were no significant differences in FeNO values according to disease severity or history of atopy (*P*>.05 for both). The same was true for the

control group, ie, no significant differences in FeNO values were obtained between the period with respiratory symptoms and recovery.

FeNO was repeated in only 29 participants during hospitalization, as the rest were lost to follow-up or their clinical condition became severe. A significant decrease in FeNO was observed in these patients (10.9 [9.1] ppb at admission vs 2.7 [2.8] ppb at the last measurement during hospitalization [*P*=.03]) (Figure S2 supplementary). All these patients were treated with systemic corticosteroids. The initial decrease is likely due to the effect of systemic corticosteroids on inducible NO synthase and is not explained by the natural progress of the infection.

Fifteen patients were admitted to the ICU (30% of all those admitted to hospital). FeNO values increased slightly during recovery compared with baseline (15.8 [14.2] ppb vs 11.1 [1.2] ppb [*P*=.12]). One patient died (initial FeNO,

Table. Demographic and Disease Characteristics Among the Patient Population^a

	Mild SARS-CoV-2 infection	SARS-CoV-2 pneumonia	SARS-CoV-2 severe pneumonia	Control group (SARS-CoV-2 -negative)	<i>P</i> Value
No. of patients	25	26	31	18	
Demographic characteristics					
Male sex	14 (56%)	21 (80.7%)	18 (58.1%)	10 (52.6%)	NS
Age, y	52.4 (15.1)	51 (14.3)	52.3 (14.7)	51.5 (14.8)	NS
Body mass index	26.9 (5.2)	26.9 (5.2)	27.2 (5.4)	26.5 (4.9)	NS
Caucasian race	16 (64%)	19 (73.1%)	18 (58.1%)	12 (63.2%)	NS
Atopy	7 (28%)	4 (15.4%)	9 (29.0%)	5 (26.3%)	NS
Allergic rhinitis	5 (20%)	4 (15.4%)	6 (19.4%)	4 (21.1%)	NS
Asthma	2 (8%)	3 (11.5%)	4 (12.9%)	4 (21.1%)	NS
No history of smoking	14 (56%)	11 (42.3%)	15 (48.4%)	12 (63.2%)	NS
Current smoker	1 (4%)	5 (19.2%)	1 (3.2%)	5 (26.3%)	NS
Ex-smoker	9 (36%)	11 (42.3%)	15 (48.4%)	2 (10.5%)	NS
Hypertension	1 (4%) ^a	8 (30.7%) ^b	7 (22.6%)	2 (10.5%)	.02
Diabetes	2 (8%)	6 (23.1%)	2 (6.4%)	1 (5.3%)	NS
Dyslipidemia	0 (0%)	5 (19.2%)	4 (12.9%)	2 (10.5%)	NS
Clinical characteristics					
Days with symptoms	7.1 (4.5)	7.2 (4.4)	7.4 (4.6)	6.9 (4.58)	NS
Fever	16 (64%)	22 (84.6%)	24 (77.4%)	6 (31.6%)	NS
Cough	22 (88%)	21 (80.7%)	27 (87.1%)	11 (57.9%)	NS
Breathlessness	7 (28%)	12 (46.1%)	25 (80.6%)	6 (31.6%)	NS
Presence of pneumonia	0 (0%)	26 (100%)	31 (100%)	2 (10.5%)	NS
Bilateral pneumonia	0 (0%)	18 (69.2%)	30 (96.7%)	1 (5.5%)	NS
Hospital admission	2 (8%) ^b	17 (65.4%) ^b	31 (100%) ^c	0 (0%) ^c	<.01
ICU admission	0 (0%)	2 (7.7%)	13 (41.9%)	0 (0%)	NS
Death	0 (0%)	0 (0%)	1 (3.2%)	0 (0%)	NS
Disease-related additional tests					
Lymphocytes/ μL	1244.1 (721.7)	1229.4 (710.4)	1201.7 (727.9)	1212.5 (853)	NS
Eosinophils/ μL	30.2 (146.4)	29.2 (146.4)	29.5 (156.7)	41.1 (173.3)	NS
FeNO at baseline, ppb	13.0 (12.4)	12.9 (12.2)	12.9 (12.9)	13.2 (12.4)	NS
Control of FeNO at recovery phase, ppb	21.6 (23.2)	20.6 (22.9)	21.3 (24.9)	21.5 (23.2)	NS
Difference between the 2 FeNO measurements, ppb	+ 8.6 (<i>P</i> =.13)	+ 7.6 (<i>P</i> =.2)	+ 8.36 (<i>P</i> =.4)	+ 8.35 (<i>P</i> =.6)	NS

Abbreviations: FeNO, fractional exhaled nitric oxide; ICU, intensive care unit; NS, nonsignificant.

^aQuantitative variables are expressed as mean (SD); qualitative variables are expressed as total number of events (percentage).

^bA significant difference was obtained for presence of hypertension between mild and moderate SARS-CoV-2 infection (*P*<.05)

^cA significant difference was obtained for hospital admission in all intergroup comparison (all *P*<.01), except between mild infection and control group (*P*=.5)

28 ppb), although the cause of the death was nonrespiratory complications.

The issue of how respiratory infections affect FeNO levels is controversial. Kharitonov et al [1] reported that FeNO increases 3-fold during clinical upper respiratory tract infections. However, the authors did not perform a viral confirmation test. Malka et al [3] demonstrated increased FeNO in children with acute viral asthma exacerbations and a positive viral nasopharyngeal PCR result, although they did not report the virus causing the disease. In contrast, Wang et al [4] showed variable FeNO levels in patients with lower respiratory tract infection depending on the virus isolated. Significantly lower values were observed for adenovirus, influenza A, parainfluenza, rhinovirus, metapneumovirus, and respiratory syncytial virus (RSV); in contrast, bocavirus infection resulted in significantly higher FeNO values. Gadish et al [7] demonstrated that FeNO levels were significantly lower during acute RSV infection despite increased FeNO production and activity in *in vitro* and animal studies. Salem et al [8] showed lower FeNO levels in patients who had recovered from SARS-CoV-2 pneumonia.

Epithelial inducible nitric oxide synthase expression is increased in some viral infections (eg, HRV, RSV, influenza A) [1,2] and in response to proinflammatory cytokines in patients with coronavirus infection. Hosts who are deficient in airway NO may have impaired respiratory antiviral defense. Martel et al [9] recently reported increased susceptibility to SARS-CoV-2 infection in patients with low airway NO levels. NO gas therapy has also been investigated as a treatment in patients with severe hypoxemia in SARS-CoV-2 infection.

To our knowledge, this is the first study to evaluate FeNO levels during acute symptomatic SARS-CoV-2 infection. The limitations are the poor definition of the possible respiratory infection in the control group, the possibility that other respiratory diseases may be a confounder, and the fact that atopic status was based on patient-reported information rather than on a confirmed diagnosis of allergy.

In summary, we found FeNO levels to be within the normal range during acute symptoms of SARS-CoV-2 infection, with some increase during the recovery phase, independently of disease severity or the patient's history of atopy. However, in patients treated with corticosteroids, a significant, more pronounced decrease in FeNO was observed during the course of the infection. These results suggest that FeNO is not a good biomarker for diagnosis or for assessment of severity or prognosis of SARS-CoV-2 infection, although more studies are necessary to confirm this hypothesis.

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Conflicts of Interest

D Betancor is supported by a Rio Hortega Research Contract from Instituto Carlos III, Ministry of Science. M Valverde-Monge has received fees for lectures from

GSK and is a member of the advisory board for Organon. JM Olaguibel reports personal fees from AstraZeneca, Mundipharma, ALK, and Eversens. J Sastre reports having served as a consultant to Thermo Fisher, MEDA, Novartis, Sanofi, Leti, Faes Farma, Mundipharma, and GSK. He has also been paid lecture fees by Novartis, GSK, Stallergenes, Leti, and Faes Farma and has received grant support for research from Thermo Fisher, Sanofi, and ALK. The remaining authors declare that they have no conflicts of interest.

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