Dear Editor

In a very recent paper by Tascini et al., the authors reported that, using oxygen-ozone (O₂–O₃) via auto-hemotherapy (O₃-AHT) in patients with mild to moderate pneumonia, statistically significant improvements occurred in 53% of O₃-AHT patients respect to controls treated with the solely best available therapy (BAT) [1]. The authors, moreover, reported that O₃-AHT treated patients showed a reduced extent in the clinical stability, yet this statement appears garbling and we would like to be elucidated about its meaning [1]. The overall impression we hold from reading this article is that Tascini et al., performed quite a pre-test, for assessing ozone effectiveness, rather than a complete therapeutic protocol on patients, following elsewhere reported methods [2]. We are persuaded that the approach used by the authors does not fit the clinical phenotypes enrolled to undergo the described therapy.

Actually, they reported that the clinical phenotypes they recruited ranged from phenotype 2 (patients with fever, chest tomography positive, presence of pulmonary consolidation area but with PO₂ > 60 mm Hg) to phenotype 4 (suspected ARDS or highly severe pneumonia to be held in sub intensive or intensive units), for whom O₃-AHT should be properly differentiated. While the authors used a single O₃-AHT approach for any phenotype, therapy protocols used by the Italian Society of Oxygen-Ozone Therapy (SIOOT) recommend, for example different routes for phenotype 2 (3 O₃-AHT/week 40–50 μg/ml on 200 ml) respect to phenotype 4 or worst (4 O₃-AHT/week or 1 O₃-AHT/day for first week, 40–50 μg/ml on 200 ml, 4 O₃-AHT/ 2nd week and 3 O₃-AHT/ 3rd week) [2]. The ability to use correctly and properly any current O₂–O₃ therapy protocol needs authors for being endowed with a full expertise about the application of O₃-AHT on COVID-19 cannot be tentatively arranged. Furthermore, authors did not describe which technology they used. The kind of device used to introduce O₂–O₃ in a subject via autohemotherapy, is crucial to warrant for the successful outcome of O₃-AHT [2]. These devices, endowed with straightforward microprocessors able to precisely tune the ratio of oxygen and ozone in the mix, are particularly sensitive to patient’s clinical conditions and adjustment is due to the validated expertise of the physician at patient’s bed [2]. Moreover, during therapy, the SatO₂% is particularly sensitive to the introduction of O₂–O₃ with O₃-AHT, whereas the authors did not report significant increase in SatO₂% during the treatment, a marker particularly crucial in assessing and tuning O₃-AHT in the patient. This information is crucial to elucidate the evidence reported by the authors, as the O₃-AHT efficacy closely depends on the correct oxygen-ozone mixture and on ozone purity and stability along time.

This may explain why results indicate a quite failing action of O₃-AHT in treated patients and this evidence is particularly caused by the absence of a therapeutic time course, i.e. a protocol using the more correct dosage of ozone and the more proper time intervals of treatments. Flaws are therefore present due both to poor statistics and lacking standardized protocols of therapy. Despite the declared difference in age and disease severity between patients selected to undergo O₃-AHT supporting BAT and patients with only BAT, the Wilcoxon signed rank test for this matching gives $p = 0.92956$ for the whole data and $p = 0.94761$ for data under the heading BGA-Admission (ref Table 1 in ref [1], suggesting that differences in the separate cohorts of patients.
were due to chance for a probability higher than 92%, i.e. that the cohorts actually belong to the same population of recruited subjects. This would mean that, aside from PaO₂/FiO₂ ratio \( (p = 0.0234) \) and SpO₂/FiO₂ ratio \( (p = 0.0202) \), patients undergoing \([O₃-AHT + BAT]\) and controls, simply treated with BAT, did not differ for any further investigated parameter upon admission. No ANCOVA, co-morbidity and confounders tests were considered to highlight if improvements were substantially due \( O₃-AHT \) therapy rather than BAT solely. Despite the authors performed a test rather than a thorough therapy protocol, some positive outcome was reported, so assessing that ozone works on COVID-19 affected subjects.

The authors reported that SIMEU hallmarks, used to set at least five clinical phenotypes, with exacerbation increasing degrees leading to suspected ARDS and sub-intensive care units, improved during hospitalization in the whole population of patients \( (p = 0.002) \) and that the observed reduction in severity was more pronounced in \( O₃-AHT \) treated patients \( (p < 0.001) \), reported as an improvement in 53% of patients treated with \( O₃-AHT \) respect to a 33% in the control cohort [1]. Ameliorations observed upon \( O₃-AHT \) therapy associated with BAT regarded only 16 on 30 patients (53.3%, from SIMEU phenotype IV/III to II) and only 25% (4 on 16) from SIMEU phenotype IV to II, whereas about 40% did not change their SIMEU phenotype (91.6% type II). Therefore, the best outcome reported, regarded only 4 patients on 16, passing from cases having a moderate to severe respiratory syndrome \( (PO₂ \text{ lower than 60 mmHg in ambient air}) \) and/or positive chest imaging for bilateral pulmonary consolidation to a mild respiratory impairment with \( PO₂ \) higher than 60 mmHg in ambient air. This quite modest improvement of COVID-19 associated pneumonia with \( O₃-AHT \), accounting for only 13% of the most outstanding results, is quite disappointing, if taking into account more encouraging previously reported effects from \( O₃-AHT \) in ICU COVID-19 positive elderly people [2]. The complete lacking of a standardized, approved protocol from the authoritative Scientific Committees in the field, made this evidence particularly biased. Actually, the way by which the authors represented their results appears anecdotal. Fundamentally with \( O₃-AHT \) it can be presumed that only that 10 patients turned from SIMEU phenotype III–II so the improvements with \( O₃-AHT \) upon BAT should regard only 6 patients on 60 (10%). This might be a good though modest result if the reader could be informed about which parameter ameliorated following \( O₃-AHT \) therapy. Unfortunately post-therapy lab data are lacking and the authors limited to referring simpler clinical phenotype classifications as markers of their therapy outcomes.

In conclusion, although the evidence reported by Tascini et al., seems to confirm a possible role of \( O₃-AHT \) therapy in COVID-19 associated pneumonia, the lack of an adequate differential therapeutic protocol for specific patients’ phenotypes and of a thorough technological description of the treatment described, makes these data particularly poor.

### Compliance with ethical standards

**Conflict of interest** The Authors state they have no conflict of interest.

**Research involving human participants and/or animals** The article does not include research involving human or animal subjects.

**Informed consent** None.

### References

1. Tascini C, Sermann G, Pagotto A, Sozio E, De Carlo C, Giacinta A, Sbrana F, Ripoli A, Castaldo N, Merelli M, Cadeo B, Macor C, De Monte A (2020) Blood ozonization in patients with mild to moderate COVID-19 pneumonia: a single centre experience. Intern Emerg Med. https://doi.org/10.1007/s11739-020-02542-6

2. Franzini M, Valdenassi L, Ricevuti G, Chirumbolo S, Depfenhart M, Bertossi D, Tirelli U (2020) Oxygen-ozone (O₂–O₃) immunocutical therapy for patients with COVID-19. Preliminary evidence reported. Int Immunopharmacol 88:106879. https://doi.org/10.1016/j.intimp.2020.106879

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