

Flora Marzia Liotti<sup>a</sup>, Brunella Posteraro<sup>b</sup>, Maria del Carmen Pereyra Boza<sup>b</sup>, Gennaro De Pascale<sup>c</sup>, Eloisa Sofia Tanzarella<sup>c</sup>, Giovanni Gherardi<sup>d</sup>, Roberta Veralli<sup>d</sup>, Maurizio Sanguinetti<sup>b</sup>, Francesco Michelangelo Turrini<sup>e</sup>

<sup>a</sup>Dipartimento di Scienze di Laboratorio e Infettivologiche, Fondazione Policlinico Universitario A. Gemelli IRCCS - Roma

<sup>b</sup>Dipartimento di Scienze Biotecnologiche di Base, Cliniche Intensivologiche e Perioperatorie, Università Cattolica del Sacro Cuore – Roma

<sup>c</sup>Dipartimento di Scienze Dell'emergenze, Anestesiologiche e Della Rianimazione, Fondazione Policlinico Universitario A. Gemelli IRCCS – Roma

<sup>d</sup>Dipartimento di Medicina e Chirurgia, Università Campus Bio-Medico di Roma – Roma

<sup>e</sup>Dipartimento di Oncologia, Università di Torino - Torino

**“Sepsis is a life-threatening condition that arises when the body’s response to an infection injures its own tissues and organs”**

## Background

Sepsis is defined as a life-threatening organ dysfunction caused by an altered host immune response to infection. We evaluated a new in-house multiplex real-time PCR assay to investigate the gene expression of pro-inflammatory and anti-inflammatory cytokines in whole-blood samples of patients with suspected sepsis. Results were assessed according to the microbiological and clinical outcomes of infection.

## Materials/methods

A prospective multicentric observational study was conducted at the Campus Biomedico and the Policlinico A. Gemelli hospitals in Rome, Italy, between April 2021 and August 2022. We included 269 patients for whom whole-blood samples were obtained at the time of sepsis suspicion. Using a multiplex real-time PCR assay developed by us, expression levels of IL-1, IL-1ra, IL-6, IL-10, IFN-gamma, or TNF-alfa genes were detected, and the beta-2 globulin gene was used as a positive control (figure 1). Ninety-eight (36.4%) patients had positive blood culture (BC) results, and 40 (14.9%) patients did not survive.

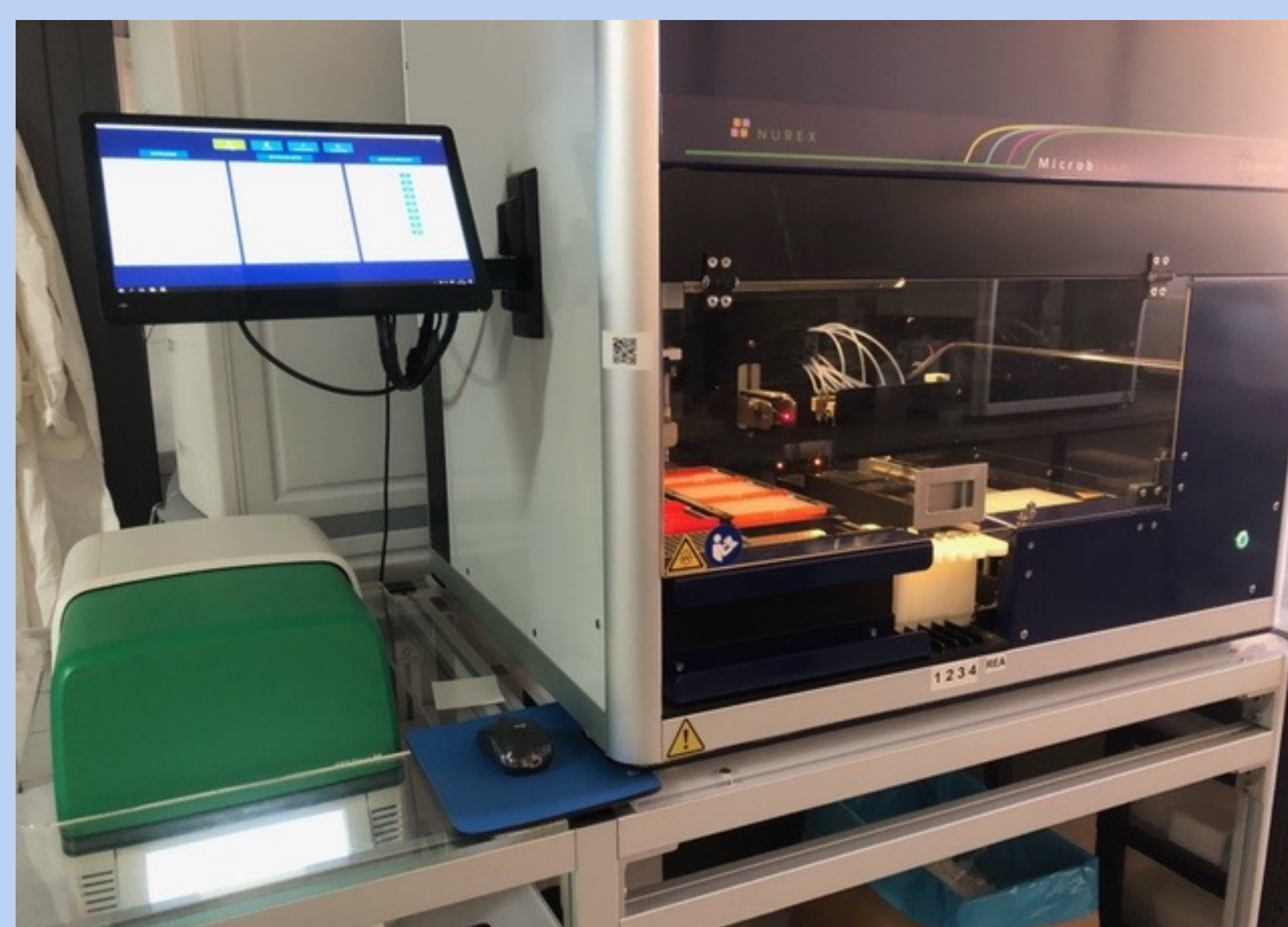
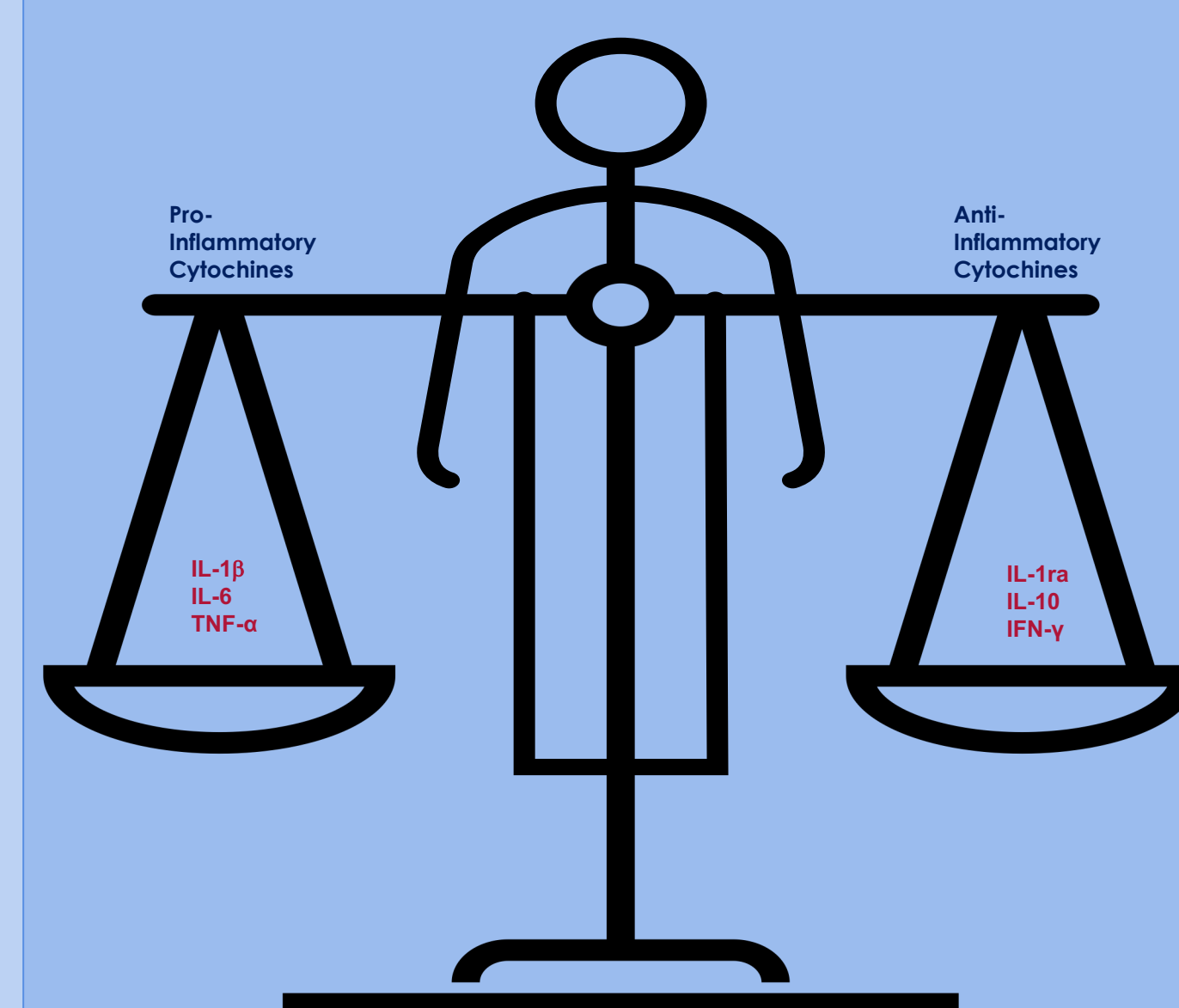


Figure1: Combination of extraction platform and amplification instrument

## Results

Overall, 195 (72.5%) of 269 samples showed a mean value of gene expression that was at least 2-fold (2.1- to 68.2-fold) higher than the baseline value for all six cytokines tested. Across these samples, IL-10 gene expression was 24.4-fold higher than the baseline value. Forty (14.9%) of 269 samples showed a mean cytokine-gene expression value that was 1.4-fold higher than the baseline value, whereas the remaining 34 (12.6%) samples had a mean cytokine-gene expression value that was 0.5-fold lower than the baseline value. Overall, gene expression levels of IL-1ra, IL-1, IL-6, IFN-gamma, and TNF-alfa were, respectively, 7.2-, 2.1-, 4.9-, 3.2-, or 4.2-fold higher than the baseline value. Samples from BC-positive patients had a mean cytokine-gene expression value that differed from the value of samples from BC-negative patients (9.4-fold and 6.8-fold higher than the baseline value, respectively;  $P=0.016$ ). Patients who survived at 28 days had a mean cytokine-gene expression value that did not differ from the value of samples from patients who did not survive (7.3-fold and 10.8-fold higher than the baseline value, respectively;  $P=0.19$ ).

Dinamic balance of pro- and anti- inflammatory cytokines



## Conclusions

Our findings show the potential of PCR-based detecting cytokine gene expression in patients with suspected sepsis. Further studies will help us to better understand these findings.

## References

1. Tsalik EL, Henao R, Montgomery JL, et al. Discriminating Bacterial and Viral Infection Using a Rapid Host Gene Expression Test. *Crit Care Med.* 2021;49(10):1651-1663.
2. Karamouzos V, Giamarellos-Bourboulis EJ, Velissaris D, Gkavogianni T, Gogos C. Cytokine production and outcome in MDR versus non-MDR gram-negative bacteraemia and sepsis. *Infect Dis (Lond).* 2021;53(10):764-771.
3. Grealy R, White M, Stordeur P, et al. Characterising cytokine gene expression signatures in patients with severe sepsis. *Mediators Inflamm.* 2013;2013:164246.
4. Chousterman BG, Swirski FK, Weber GF. Cytokine storm and sepsis disease pathogenesis. *Semin Immunopathol.* 2017;39(5):517-528. doi:10.1007/s00281-017-0639-8
5. Mierzczyńska-Pasierb M, Lipińska-Gediga M. Sepsis diagnosis and monitoring - procalcitonin as standard, but what next?. *Anaesthesiol Intensive Ther.* 2019;51(4):299-305.