Monocyte and macrophage phenotypes: a look beyond systemic sclerosis. Response to: 'M1/M2 polarisation state of M-CSF blood-derived macrophages in systemic sclerosis' by Lescoat *et al*

We read with interest the new data from Lescoat *et al* and their interesting comments concerning our research letter on the identification of circulating cells coexpressing M1 and M2 phenotype markers in patients affected by systemic sclerosis (SSc) compared with healthy subjects (HSs).[1 2]

In their study, Lescoat *et al* evaluated the mean of fluorescence intensity of specific markers of M1 and M2 phenotypes (CD80, CD206, CD204, CD163, CD169 and CD200R1) in macrophage colony-stimulating factor (M-CSF) resting blood monocyte-derived-macrophages (MDMs) from HSs exposed to in vitro stimuli aiming to polarise them towards the M1, M2a and M2c phenotypes. The markers were also evaluated on the cell surface of the MDMs obtained from 11 to 16 SSc patients not treated with glucocorticoids and compared with those obtained from 13 HSs.[1] The important conclusion highlighted is cultured circulating MDMs from patients with SSc showcells with a mixed M1/M2 signature, supporting our results.[2]

We will soon be able to complement these data with the clinical implications seen in a new study that is going to demonstrate, in patients with SSc, how the higher percentages of circulating cells showing a mixed M1/M2 phenotype correlate significantly with important SSc clinical complications, such as functional and structural lung damage (data under publication).

Lescoat *et al* appropriately asserted that the results showing a mixed phenotype of circulating monocyte/macrophage cells in patients with SSc might arise both methodological and conceptual questions with respect to the M1/M2 definition.

Lescoat's results further highlighted how macrophages can evolve to exhibit characteristics that are shared by more than one macrophage population, similarly to secondary colours in a colour wheel, as postulated by Mosser and Edwards.[3]

Although it is true that our study was conducted on circulating and theoretically less differentiated cells, it is also possible to hypothesise that the in vitro differentiation of monocytes through the stimulation with M-CSF as well as GM-CSF probably does not fully reproduce the process induced by the in vivo microenvironmental signals.[4 5] Also, the comparison with cultured macrophages differentiated from human cell lines, such as primarily PMA-treated THP-1 cell line, would be even less accurate.[6]

Furthermore, the higher percentages of differentiated and activated myeloid-derived circulating cells in patients with SSc compared with HSs make part of the most interesting results of our research. Our data and those from Lescoat *et al* could in any case coexist, without being in contrast with the different findings observed at tissue level, that is, in internal organs affected by SSc, since the different tissues are able to direct the inflammatory response in a proinflammatory direction or towards resolution and repair, determining the expression of a more polarised and stable cell phenotype in the periphery (ie, M1 and/or M2).[7]

Nevertheless, the more differentiated phenotype observed at the tissue level could not mirror the one observed in circulating monocyte/macrophage precursors or that derived from in vitro MDMs. Additionally, it was very recently demonstrated that different cell phenotypes could be observed in different tissues especially in SSc and, particularly for innate immune cells, given their plasticity.[4 5 8]

Finally, the acquisitions on monocyte/macrophage polarisation have shown at least that the approach based on the evaluation of single or few markers for the determination of a cell signature is no longer conceivable. In fact, the strong interest currently found in the contribution of the innate immune cells to pathogenic processes should probably be directed towards the clarification of the relationships between the phenotype of circulating cells and that of more differentiated ones observed in peripheral tissues and to a wider phenotype study and definition.

The results described in our study and that presented by Lescoat *et al* contributed to identify possible new cell players involved in the pathophysiology of SSc.

In accordance with Behmoaras and Petretto, the question regarding whether and how the circulating mixed M1/M2 cells described in our study and/or the MDMs investigated in vitro by Lescoat *et al* could reflect the context-specific activation of macrophages in the different SSc tissues (ie, lung and skin) remains open and matter of our further research.[4]

Starting from these results, the functional role of mixed M1/M2 cells in the pathogenesis of SSc and in other fibrotic diseases, as well as the possible effects of modulators and the related clinical complications, should be better addressed.[1 5 9] These considerations on the innate immune cell plasticity could apply to the contribution to SSc pathogenesis and to other fibrotic diseases and pathological or even physiological conditions.[7]

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Handling editor Josef S Smolen

Funding The authors have not declared a specific grant for this research from any funding agency in the public, commercial or not-for-profit sectors.

Competing interests None declared.

Patient consent Not required.

Provenance and peer review Commissioned; internally peer reviewed.

MC, ACT and SS contributed equally.

To cite Cutolo M, Trombetta AC, Soldano S. *Ann Rheum Dis* 2019;**78**:e128.

Received 7 September 2018
Accepted 7 September 2018
Published Online First 29 September 2018

Linked

► https://doi.org/10.1136/annrheumdis-2018-214333

Ann Rheum Dis 2019;**78**:e128. doi:10.1136/annrheumdis-2018-214371

REFERENCES

1 Lescoat A, Ballerie A, Jouneau S. M1/M2 polarization state of M-CSF blood-derived macrophages in systemic sclerosis. *Ann Rheum Dis* 2019;78:e127–e127.
2 Soldano S, Trombetta AC, Contini P, *et al*. Increase in circulating cells coexpressing M1 and M2 macrophage surface markers in patients with systemic sclerosis. *Ann Rheum Dis* 2018;77:1842–5.
3 Mosser DM, Edwards JP. Exploring the full spectrum of macrophage activation. *Nat Rev Immunol* 2008;8:958–69.
4 Behmoaras J, Petretto E. Cell function in disease: there are more than two parties at play. *Ann Rheum Dis* 2019;78:e20–e20. 10.1136/annrheumdis-2018-213121.
5 Moreno-Moral A, Bagnati M, Koturan S, *et al*. Changes in macrophage transcriptome associate with systemic sclerosis and mediate *GSDMA* contribution to disease risk. *Ann Rheum Dis* 2018;77:596–601.
6 Forrester MA, Wassall HJ, Hall LS, *et al*. Similarities and differences in surface receptor expression by THP-1 monocytes and differentiated macrophages polarized using seven different conditioning regimens. *Cell Immunol* 2018;332:58–76.
7 Matzinger P, Kamala T. Tissue-based class control: the other side of tolerance. *Nat Rev Immunol* 2011;11:221–30.
8 Taroni JN, Greene CS, Martyanov V, *et al*. A novel multi-network approach reveals tissue-specific cellular modulators of fibrosis in systemic sclerosis. *Genome Med* 2017;9:27.
9 Soldano S, Pizzorni C, Paolino S, *et al*. Correction: alternatively activated (M2) macrophage phenotype Is inducible by endothelin-1 in cultured human macrophages. *PLoS One* 2017;12:e0175238.