COMMENTS FROM THE REFEREE

General assessment: This is a well-written, thoroughly referenced, well-organized and comprehensive review article on a subject which is appropriate to the World Journal of Stem Cells. I found it to be very useful in covering the large recent literature on this theme, and believe even the specialists on mesenchimal stem cells may profit from its coverage of an important subset of MSC available through dental and craniofacial surgery. The following specific points are raised to suggest improvements on the immunological aspects of the text.

AUTHORS' ANSWER

We are thankful to the Reviewer for the evaluation and critical comments on our manuscript.

SPECIFIC COMMENT: 1.

The potent immunomodulatory cytokine TGF-β is continuously produced by MSCs, and its production can be enhanced by other anti-inflammatory factors, such as IL-4 and IL-13 [20]. IL-10 is an anti-inflammatory cytokine that can be produced either by MSCs or MSC-instructed immune cells [21].
Comment: many studies show that IL-4 and IL-13 are essential components of the immediate hypersensitivity reaction (IgE- and IgG1-mediated); furthermore, administration of IL-13 alone can induce a severe asthma-like disease in mice [M. Wills-Karp and colleagues have extensively documented this phenomenon]. Since these are severe inflammatory conditions, although of a specialized type, it is inappropriate to deduce that IL-4 and IL-13 are "anti-inflammatory factors". This view, which is widespread, originates in the misconception that "inflammation" is synonymous with TH1-dependent cellular immune reactions. Instead, TGF-β, IL-4 and IL-13 are all immunomodulatory cytokines, which means that they modify immune response patterns, opposing some forms of immune-mediated inflammation while favoring others. In the case of TGF-β, it can be strongly proinflammatory when associated with IL-6, by promoting TH17 cell differentiation. All three immunomodulatory cytokines contrast with IL-10, which is, for most experimental settings, clearly anti-inflammatory and immunosuppressive.

AUTHORS' ANSWER

We appreciate this comment. In the revised version, we refer IL-4 and IL-13 as immunomodulatory and not as anti-inflammatory cytokines.

SPECIFIC COMMENT: 2.

In most studies, dental MSCs have been co-cultured with peripheral blood mononuclear cells (PBMCs), followed by the analysis of specific markers and/or functional characteristics of different immune cell subsets. These experimental approaches have some advantages and limitations.
Comment: In my view, the authors did not address the major limitation of the co-culture approach, which is the artificiality of the system. It is true that many controls can be carried out in such co-culture studies, but the major criticism still applies, that these models do not duplicate any known in vivo interaction. For instance, a mixture of circulating blood mononuclear cells is not likely to interact in vivo with mesenchimal stem cells, which live in different extravascular compartments. Again,

Concanavalin- A or otherwise (PHA-, anti-CD3/CD28 antibodies-) activated PBMC are hardly representative of physiologically activated cells. This limitation is often overlooked and a review article should not omit this fact.

AUTHORS' ANSWER

Thank you for pointing out this issue. We have mentioned the limitations of co-culture approaches in the revised version of our manuscript.

SPECIFIC COMMENT: 3.

Although DPSCs, like other MSCs, are immunotolerant, they can sometimes be susceptible to NK cell-mediated cytotoxicity [33]. Comment: This sentence is difficult to understand. An organism which is immunotolerant shows specific tolerance to a given antigen to which it would otherwise react strongly and specifically. By extension of this concept from organisms to cells, one might say that T cells from tolerant donors may have decreased or absent reactivity agains some antigen to which they would normally react strongly. I cannot see how mesenchimal stem cells, which are not able to react specifically to antigens, can become tolerant in this sense. At any rate, that would not have an obvious connection with the susceptibility to the lytic action of NK cells, which do not specifically recognize antigens, but other activator or inhibitor ligands.

AUTHORS' ANSWER

We fully agree that it is not correct to compare immunotolerance and susceptibility to NK-mediated lysis. The sentence mentioned by the Reviewer was revised and it sounds now as "DPSCs can sometimes be susceptible to NK cell-mediated cytotoxicity.[33]"