

available and accessible to broad patient populations due to their undetectable levels of major histocompatibility complex class II (MHC-II) molecules and low levels of MHC-I.

There are some preclinical data supporting efficacy of MSCs in AD. In animal models, MSCs cross the BBB, promote neurogenesis, inhibit β -amyloid deposition and promote clearance, reduce apoptosis, promote hippocampal neurogenesis, improve dendritic morphology, and improve behavioral and spatial memory performance [18-20]. These beneficial effects were associated with decreased inflammation, increased A β -degrading factors and A β clearance, decreased hyperphosphorylated tau, and elevated alternatively activated microglial markers. These benefits appear, at least in part, due to A β -induced MSC release of chemoattractants that recruit alternative microglia into the brain to reduce A β deposition [21]. MSCs have been reported to be effective in young AD-model mice prior to A β accumulations, leading to significant decreases in cerebral A β deposition, and a significant increase in expression of pre-synaptic proteins [22]. Impressively, these effects were sustained for at least 2 months, and suggest MSCs could potentially be effective as an interventional therapeutic in prodromal AD.

Accordingly, the application seeks to not only provide methods of treatment for AD wherein the methods comprise the use of compositions containing MSCs, but this application also seeks to provide methods that can accurately measure the potential safety of MSCs and evaluate their efficacy in the alleviation of AD symptoms in subjects in need thereof.

SUMMARY

An objective of the present application is to provide a use of allogeneic mesenchymal stem cells (MSCs) in the manufacture of a medicament for alleviating the symptoms of Alzheimer's disease (AD) in a subject in need thereof. In another aspect, it is referred to a use of allogeneic mesenchymal stem cells (MSCs) in the manufacture of a medicament for treating Alzheimer's disease (AD) or inhibiting AD disease progression. Also disclosed herein are methods of treatment or alleviation for AD that comprise administering a therapeutic amount of allogeneic MSCs to a subject in need thereof to alleviate the symptoms and/or treat the progression of AD. Another example of the present application is to provide novel biomarkers for diagnosing and evaluating the progression of AD and the effectiveness of the treatment methods. These biomarkers may be the change in the size in areas of a patient's brain, such as the amygdala, cortical nucleus, the hippocampus, hippocampal subregions, and/or the corticoamygdaloid transition.

In some example, the novel biomarkers for diagnosing and evaluating the progression of AD may be a change in a cytokine's concentration, wherein the cytokine may be IL-4, IL-6, IL-8, IL-10, IL-12p70, IL-17, sIL-2R α or combinations thereof. In preferred

example, the cytokine's concentration is increased in the serum, plasma, cerebral spinal fluid or blood of the subject in need thereof suffering from AD symptoms after administration of allogenic MSCs to said subject. The cytokine concentration increase can range from 0% to 10%, 0.5% to 10%, 1.0% to 10%, 3% to 10%, 5% to 10%, 7% to 10%, greater than 0% to less than or equal to 10%, 10% to 50%, 20% to 50%, 30% to 50% or greater than 50%. In preferred example, the cytokine concentration is increased to a stable concentration level wherein the concentration does not decline more than 0% to 10%, 0% to 5% or 0% to 1% once it has reached and maintained a concentration level that is different from the concentration level before administration of MSCs to the subject in need thereof.

In other example, the novel biomarkers for diagnosing and evaluating the progression of AD may be a change in a neuronal-related molecule or peptide's concentration, wherein the neuronal signaling molecule or peptide may be tau, phospho-tau, A β -38, A β -40, A β -42, NFL or combinations thereof. In preferred example, the concentration of A β -38, A β -40 or A β -42 is increased in the serum, plasma or blood of the subject in need thereof suffering from AD symptoms after administration of allogenic MSCs to said subject. The A β -38, A β -40 or A β -42 concentration increase can range from 0% to 10%, 0.5% to 10%, 1.0% to 10%, 3% to 10%, 5% to 10%, 7% to 10%, greater than 0% to less than or equal to 10%, 10% to 50%, 20% to 50%, 30% to 50% or greater than 50%. In preferred example, the A β -38, A β -40 or A β -42 concentration is increased to a stable concentration level wherein the concentration does not decline more than 0% to 10%, 0% to 5% or 0% to 1% once it has reached and maintained a concentration level that is different from the concentration level before administration of MSCs to the subject in need thereof.

In other example, the concentration of tau, phospho-tau, or NFL is decreased in the serum, plasma, cerebral spinal fluid or blood of the subject in need thereof suffering from AD symptoms after administration of allogenic MSCs to said subject. The tau, phospho-tau, or NFL concentration decrease can range from 0% to 10%, 0.5% to 10%, 1.0% to 10%, 3% to 10%, 5% to 10%, 7% to 10%, greater than 0% to less than or equal to 10%, 10% to 50%, 20% to 50%, 30% to 50% or greater than 50%. In preferred example, the tau, phospho-tau, or NFL concentration is decreased to a stable concentration level wherein the concentration does not increase more than 0% to 10%, 0% to 5% or 0% to 1% once it has reached and maintained a concentration level that is different from the concentration level before administration of MSCs to the subject in need thereof.

In other example, the novel biomarkers for diagnosing and evaluating the progression of AD may be a change in an inflammation signaling molecule's concentration, wherein the inflammation signaling molecule may be pro-BNP, TNF- α , or combinations thereof. In preferred example, the concentration of TNF- α or pro-BNP is decreased in the serum, plasma, cerebral spinal fluid or blood of the subject in need thereof suffering from AD symptoms after administration of allogenic MSCs to said subject. The TNF- α or pro-BNP concentration decrease can range from 0% to 10%, 0.5% to 10%, 1.0% to 10%, 3% to 10%, 5% to 10%, 7% to 10%, greater than 0% to less than or equal to 10%, 10% to 50%, 20% to 50%, 30% to 50% or greater than 50%. In preferred example, the TNF- α or pro-BNP concentration is decreased to a stable concentration level wherein the concentration does not increase more than 0% to 10%, 0% to 5% or 0% to 1% once it has reached and maintained a concentration level that is different from the concentration level before administration of MSCs to the subject in need thereof.

In some example, the novel biomarkers for diagnosing and evaluating the progression of AD may be a change in VEGF concentration and other vascular-related biomarkers. In preferred example, the concentration of VEGF is increased in the serum, plasma, cerebral spinal fluid or blood of the subject in need thereof suffering from AD symptoms after administration of allogenic MSCs to said subject. The VEGF concentration increase can range from 0% to 10%, 0.5% to 10%, 1.0% to 10%, 3% to 10%, 5% to 10%, 7% to 10%, greater than 0% to less than or equal to 10%, 10% to 50%, 20% to 50%, 30% to 50% or greater than 50%. In preferred example, the VEGF concentration is increased to a stable concentration level wherein the concentration does not decline more than 0% to 10%, 0% to 5% or 0% to 1% once it has reached and maintained a concentration level that is different from the concentration level before administration of MSCs to the subject in need thereof.

The allogenic MSCs may be LOMECCEL-BTM cells, which is a Longeveron formulation of allogenic human mesenchymal stem cells. Further uses and preparation of useful stem cells, including LOMECCEL-BTM brand mesenchymal cells, may be found in the following United States Patent Application Publications, all of which are incorporated by reference herein: US20190038742A1; US20190290698A1; and US20200129558A1.

CLAIMS

1. Use of allogeneic mesenchymal stem cells (MSCs) in the manufacture of a medicament for alleviating the symptoms of Alzheimer's disease (AD) in a subject in need thereof.
2. Use of allogeneic mesenchymal stem cells (MSCs) in the manufacture of a medicament for treating Alzheimer's disease (AD) or inhibiting AD disease progression.
3. The use according to claims 1 or 2, wherein the use further comprises that the concentration of a biomarker or biomarkers in the subject suffering from symptoms of AD to be measured *ex vivo* before and after the administration of the allogenic MSCs.
4. The use according to any one of claims 1-3, wherein the use further comprises that the cognitive function of the subject suffering from symptoms of AD to be measured *ex vivo* before and after administration of the allogenic MSCs.
5. The use according to any one of claims 3-4, wherein the biomarkers comprise cytokines selected from the group consisting of IL-4, IL-6, IL-8, IL-10, IL-12p70, IL-17, sIL-2R α or combinations thereof.
6. The use according to claim 5, wherein the cytokine's concentration is increased in the serum, plasma, cerebral spinal fluid or blood of the subject in need thereof after administration of the allogenic MSCs.
7. The use according to claim 5, wherein the cytokine concentration increase is from 0.5% to 10%, 5% to 10%, 10% to 50%, or greater than 50%.

8. The use according to any one of claims 3-7, wherein the biomarkers further comprise neuronal-related molecules or peptides selected from the group consisting of tau, phospho-tau, A β -38, A β -40, A β -42, NFL or combinations thereof.
9. The use according to claim 8, wherein the concentration of A β -38, A β -40 or A β -42 is increased in the serum, plasma, or blood of the subject in need thereof after administration of the allogenic MSCs.
10. The use according to claim 9, wherein the concentration of A β -38, A β -40 or A β -42 is increased from 0.5% to 10%, 5% to 10%, 10% to 50%, or greater than 50%.
11. The use according to claim 8, wherein the concentration of tau, phospho-tau or NFL is decreased in the serum, plasma, cerebral spinal fluid or blood of the subject in need thereof after administration of the allogenic MSCs.
12. The use according to claim 11, wherein the concentration of tau, phospho-tau or NFL is decreased from 0.5% to 10%, 5% to 10%, 10% to 50%, or greater than 50%.
13. The use according to any one of claims 3-12, wherein the biomarkers further comprise inflammation signaling molecules such as pro-BNP, TNF- α , or combinations thereof.
14. The use according to claim 13, wherein the concentration pro-BNP or TNF- α is decreased in the serum, plasma, cerebral spinal fluid or blood of the subject in need thereof after administration of the allogenic MSCs.
15. The use according to claim 14, wherein the concentration of pro-BNP or TNF- α is decreased from 0.5% to 10%, 5% to 10%, 10% to 50%, or greater than 50%.

16. The use according to any one of claims 3-15, wherein the biomarkers further comprise VEGF.

17. The use according to claim 16, wherein the concentration of VEGF is increased in the serum, plasma, cerebral spinal fluid or blood of the subject in need thereof after administration of the allogeneic MSCs.

18. The use according to claim 17, wherein the concentration of VEGF is decreased from 0.5% to 10%, 5% to 10%, 10% to 50%, or greater than 50%.

19. The use according to any one of claims 3-18, wherein the use further comprises that the change in the size of areas in the subject's brain is to be determined after administration of the allogeneic MSCs.

20. The use according to claim 19, wherein the areas in the subject's brain that change in size after administration of the allogeneic MSCs are selected from the group consisting of the amygdala, cortical nucleus, the hippocampus, hippocampal subregions, and/or the corticoamygdaloid transition.

21. The use according to any one of claims 3-20, wherein the use further comprises that if a change occurs is to be determined in the corticoamygdaloid transition of the subject after administration of the allogeneic MSCs.

22. The use according to any one of claims 3-21, wherein the medicament comprises 20×10^6 MSCs.

23. The use according to any one of claims 3-21, wherein the medicament comprises 100×10^6 MSCs.

24. The use according to any one of claims 3-23, wherein the use further comprises that the cerebral spinal fluid of the subject is to be examined *ex vivo* before and after administration of the allogeneic MSCs.