response in a non-responding subject, comprising administering to the subject concurrently or sequentially the vaccine and an adjuvant in immunoprotective amounts, wherein the adjuvant is a population of isolated allogeneic human mesenchymal stem cells, and kits and uses related thereto. In one aspect, the present disclosure refers to the use of a composition comprising a vaccine and an immunoprotective amount of an adjuvant, the adjuvant being a population of isolated homogenous allogenic human mesenchymal stem cells in the manufacture of a medicament for enhancing a human subject's immune response to the vaccine or inducing an immune response in a human subject whose immune system did not previously react to the vaccine administered, wherein the human subject exhibits symptoms of aging frailty, the human mesenchymal stem cells are not genetically manipulated and wherein the vaccine comprises: (i) one or more inactivated viruses; (ii) one or more live, attenuated viruses; or (iii) one or more antigenic polypeptides selected from the group consisting of influenza hemagglutinin 1 (HA1), hemagglutinin 2 (HA2), influenza neuraminidase (NA), Lassa virus (LASV) glycoprotein 1 (gp1), LASV glycoprotein 2 (gp2), LASV nucleocapsid-associated protein (NP), LASV L protein, LASV Z protein, SARS virus S protein, Ebola virus GP2, measles virus fusion 1 (F1) protein, HIV-1 transmembrane (TM) protein, HIV-1 glycoprotein 41 (gp41), HIV-1 glycoprotein 120 (gp120), hepatitis C virus (HCV) envelope glycoprotein 1 (E1), HCV envelope glycoprotein 2 (E2), HCV nucleocapsid protein (p22), West Nile virus (WNV) envelope glycoprotein (E), Japanese encephalitis virus (JEV) envelope glycoprotein (E), yellow fever virus (YFV) envelope glycoprotein (E), tick-borne encephalitis virus (TBEV) envelope glycoprotein (E), hepatitis G virus (HGV) envelope glycoprotein 1 (E1), respiratory syncytial virus (RSV) fusion (F) protein, herpes simplex virus1 (HSV-1) gD protein, HSV-1 gG protein, HSV-2 gD protein, HSV-2 gG protein, hepatitis B virus (HBV) core protein, and Epstein-Barr virus (EBV) glycoprotein 125 (gp125).

[00014] In one example of the disclosure, the subject is a human. In another example of the disclosure, the subject is a human who exhibits symptoms of aging frailty. In another example of the disclosure, the subject is a human who exhibits inflammaging.

[00015] In one example of the disclosure, the mesenchymal stem cells are bone marrow-derived mesenchymal stem cells. In one example of the disclosure, the mesenchymal stem cells do not express STRO-1. In another example of the disclosure, the mesenchymal stem cells do not express CD45. In another example of the disclosure, the mesenchymal stem cells do not express fibroblast surface markers or have a fibroblast morphology. In another example of the disclosure, the mesenchymal stem cells are not genetically manipulated.

[00016] In one example of the disclosure, the vaccine is monovalent. In another example of the disclosure, the vaccine is multivalent. In one example of the disclosure, the vaccine comprises one or more inactivated viruses. In a further example, the one or more inactivated viruses are selected from the group consisting of adenoviruses, picornaviruses, papillomaviruses, polyomaviruses, hepadnaviruses, parvoviruses, pox viruses, Epstein-Barr virus, cytomegalovirus (CMV), herpes

viruses, roseolovirus, varicella zoster virus, filoviruses, paramyxoviruses, orthomyxoviruses, rhabdoviruses, arenaviruses, coronaviruses, human enteroviruses, hepatitis A virus, human rhinoviruses, polio virus, retroviruses, rotaviruses, flaviviruses, hepaciviruses, togaviruses, and rubella virus. In a further example, the vaccine comprises an inactivated orthomyxovirus. In a further example, the vaccine comprises an inactivated influenza virus. In one example of the disclosure, the vaccine comprises one or more live, attenuated viruses. In a further example, the one or more attenuated viruses are selected from the group consisting of adenoviruses, picornaviruses, papillomaviruses, polyomaviruses, hepadnaviruses, parvoviruses, pox viruses, Epstein-Barr virus, cytomegalovirus (CMV), herpes viruses, roseolovirus, varicella zoster virus, filoviruses, paramyxoviruses, orthomyxoviruses, rhabdoviruses, arenaviruses, coronaviruses, human enteroviruses, hepatitis A virus, human rhinoviruses, polio virus, retroviruses, rotaviruses, flaviviruses, hepaciviruses, togaviruses, and rubella virus.

[00017] In one example of the disclosure, the adjuvant is administered before the vaccine. In a further example, the adjuvant is administered at least 1 week before the vaccine is administered. In a further example, the adjuvant is administered at least 2 weeks before the vaccine is administered. In a further example, the adjuvant is administered at least 3 weeks before the vaccine is administered. In a further example, the adjuvant is administered at least 4 weeks before the vaccine is administered. In another example of the disclosure, the adjuvant is administered at least yearly for long-term enhancement of vaccine response.

[00018] In one example of the disclosure, the adjuvant and the vaccine are administered parenterally. In one example, the adjuvant is administered systemically. In one example of the disclosure, the adjuvant is administered by infusion or direct injection. In one example of the disclosure, the adjuvant is administered intravenously, intraarterially, or intraperitoneally. In a further example, the adjuvant is administered intravenously. In one example of the disclosure, the vaccine is administered intramuscularly, intravenously, intraarterially, intraperitoneally, subcutaneously, intradermally, orally, or intranasally. In a further example, the vaccine is administered intramuscularly.

[00019] In one example of the disclosure, the adjuvant is administered at a dose of about $20x10^6$ mesenchymal stem cells. In another example of the disclosure, the adjuvant is administered at a dose of about $100x10^6$ mesenchymal stem cells. In another example of the disclosure, the adjuvant is administered at a dose of about $200x10^6$ mesenchymal stem cells.

[00020] In one example of the disclosure, the mesenchymal stem cells are obtained from a human donor and a step of MHC matching of the human donor to the subject is not employed prior to the administration of the vaccine and adjuvant to the subject.

[00021] In one example of the disclosure, as a result of the disclosed methods of enhancing a subject's immune response to a vaccine or of inducing an immune response in a non-responding

subject, comprising administering to the subject concurrently or sequentially the vaccine and an adjuvant in immunoprotective amounts, wherein the adjuvant is a population of isolated allogeneic human mesenchymal stem cells, the intracellular TNF-α expression in the B cells of the subject decreases by at least two-fold as compared to the TNF-α expression levels in B cells of the subject prior to administration of the adjuvant. In another example of the disclosure, the $CD4^+$:$CD8^+$ T cell ratio in the subject increases by at least two-fold as compared to the $CD4^+$:$CD8^+$ T cell ratio in the subject prior to administration of the adjuvant. In another example of the disclosure, the number of switched memory B cells in the subject increases by at least two-fold as compared to the number of switched memory B cells in the subject prior to administration of the adjuvant. In another example of the disclosure, the number of exhausted B cells in the subject decreases by at least two-fold as compared to the number of exhausted B cells in the subject prior to administration of the adjuvant. In another example, the disclosure concerns a method of inducing an immune response in a subject comprising administering to the subject concurrently or sequentially a vaccine and an adjuvant in immunoprotective amounts, wherein the adjuvant is a population of isolated allogeneic human mesenchymal stem cells, and further wherein the intracellular TNF-α expression in B cells of the subject decreases by at least two-fold as compared to the TNF-α expression levels in B cells of the subject prior to administration of the adjuvant. The disclosure also concerns a method of inducing an immune response in a subject comprising administering to the subject concurrently or sequentially a vaccine and an adjuvant in immunoprotective amounts, wherein the adjuvant is a population of isolated allogeneic human mesenchymal stem cells, and further wherein the CD4+:CD8+ T cell ratio in the subject increases by at least two-fold as compared to the CD4+:CD8+ T cell ratio in the subject prior to administration of the adjuvant. The disclosure further concerns a method of inducing an immune response in a subject comprising administering to the subject concurrently or sequentially a vaccine and an adjuvant in immuoprotective amounts, wherein the adjuvant is a population of isolated allogeneic human mesenchymal stem cells, and further, wherein the number of switched memory B cells in the subject increases by at least two-fold as compared to the number of switched memory B cells in the subject prior to administration of the adjuvant. The disclosure still further concerns a method of inducing an immune response in a subject comprising administering to the subject concurrently or sequentially a vaccine and an adjuvant in immuoprotective amounts, wherein the adjuvant is a population of isolated allogeneic human mesenchymal stem cells, and further wherein the number of exhausted B cells in the subject decreases by at least two-fold as compared to the number of exhausted B cells in the subject prior to administration of the adjuvant.

[00022] In another example, the disclosure relates to a kit having at least two containers, comprising: a vaccine in a first container and an adjuvant in a second container, wherein the adjuvant is a population of isolated allogeneic human mesenchymal stem cells. In one example of the disclosure, the mesenchymal stem cells of the kit are cryopreserved. In one

example of the disclosure, the kit further comprises dilution buffer in a third container. The vaccines and the mesenchymal stem cells present in the kit may be any of the vaccines or allogeneic human mesenchymal stem cells disclosed herein.

BRIEF DESCRIPTION OF THE DRAWING FIGURES

[00023] FIG. 1 shows RT-PCR for E47 and GAPDH from five subjects.

[00024] FIG. 2 shows AID mRNA expression correlated to E47 expression. Reproduced from Figure 3 of Frasca, D. *et al., J. Immunol. 180(8)*:5283-5290 (2008).

[00025] FIG. 3 shows the % of switched B cells as measured by flow cytometry. An increase in the % of switched memory B cells was observed in subjects who received $20x10^6$, $100x10^6$, or $200x10^6$ mesenchymal stem cells at 6 months post-mesenchymal stem cell infusion as compared to the baseline measurement.

[00026] FIG. 4 shows the % of switched memory B cells and exhausted B cells as measured by flow cytometry. The % of switched memory B cells and exhausted B cells was measured in subjects who received a second mesenchymal stem cell infusion one year after their first mesenchymal stem cell infusion.

[00027] FIGS. 5A-5D show T cell concentrations as measured by flow cytometry. FIGS. 5A-5D show that there is no T cell activation (rejection) induced by the allogeneic mesenchymal stem cells at any dose. CD69 is an early marker of T cell activation. CD25 is a late/chronic marker of T cell activation.

[00028] FIG. 6 shows $CD4^+$:$CD8^+$ T cell ratios, calculated from flow cytometry measurements. An improvement in the $CD4^+$:$CD8^+$ T cell ratio (Immune Risk Phenotype) was observed in subjects who received a second mesenchymal stem cell infusion one year after their first mesenchymal stem cell infusion.

[00029] FIGS. 7A and 7B show downregulation of intracellular TNF-α in B cells by flow cytometry after mesenchymal stem cell infusion. "Base line" refers to flow cytometry measurements taken at baseline. "3 months" refers to flow cytometry measurements taken 3 months after subjects received a second mesenchymal stem cell infusion one year after their first mesenchymal stem cell infusion. "FSC-A" is forward-scattered light and is proportional to cell-surface area or size. "SSC-A" is side-scattered light and is proportional to cell

WHAT IS CLAIMED IS:

1. Use of a composition comprising a vaccine and an immunoprotective amount of an adjuvant, the adjuvant being a population of isolated homogenous allogeneic human mesenchymal stem cells, in the manufacture of a medicament for enhancing a human subject's immune response to the vaccine or inducing an immune response in a human subject whose immune system did not previously react to the vaccine administered, wherein the human subject exhibits symptoms of aging frailty, the human mesenchymal stem cells are not genetically manipulated and wherein the vaccine comprises: (i) one or more inactivated viruses; (ii) one or more live, attenuated viruses; or (iii) one or more antigenic polypeptides selected from the group consisting of influenza hemagglutinin 1 (HA1), hemagglutinin 2 (HA2), influenza neuraminidase (NA), Lassa virus (LASV) glycoprotein 1 (gp1), LASV glycoprotein 2 (gp2), LASV nucleocapsid-associated protein (NP), LASV L protein, LASV Z protein, SARS virus S protein, Ebola virus GP2, measles virus fusion 1 (F1) protein, HIV-1 transmembrane (TM) protein, HIV-1 glycoprotein 41 (gp41), HIV-1 glycoprotein 120 (gp120), hepatitis C virus (HCV) envelope glycoprotein 1 (E1), HCV envelope glycoprotein 2 (E2), HCV nucleocapsid protein (p22), West Nile virus (WNV) envelope glycoprotein (E), Japanese encephalitis virus (JEV) envelope glycoprotein (E), yellow fever virus (YFV) envelope glycoprotein (E), tick-borne encephalitis virus (TBEV) envelope glycoprotein (E), hepatitis G virus (HGV) envelope glycoprotein 1 (E1), respiratory syncytial virus (RSV) fusion (F) protein, herpes simplex virus1 (HSV-1) gD protein, HSV-1 gG protein, HSV-2 gD protein, HSV-2 gG protein, hepatitis B virus (HBV) core protein, and Epstein-Barr virus (EBV) glycoprotein 125 (gp125).

2. The use of claim 1, wherein the human subject exhibits inflammaging.

3. The use of claim 1, wherein the mesenchymal stem cells do not express STRO-1.

4. The use of claim 1, wherein the mesenchymal stem cells do not express CD45.

5. The use of claim 1, wherein the mesenchymal stem cells do not express fibroblast surface markers or have a fibroblast morphology.

6. The use of claim 1, wherein the vaccine is multivalent.

7. The use of claim 1, wherein the one or more inactivated viruses are selected from the group consisting of adenoviruses, picornaviruses, papillomaviruses, polyomaviruses, hepadnaviruses, parvoviruses, pox viruses, Epstein-Barr virus, cytomegalovirus (CMV), herpes viruses, roseolovirus, varicella zoster virus, filoviruses, paramyxoviruses, orthomyxoviruses, rhabdoviruses, arenaviruses, coronaviruses, human enteroviruses, hepatitis

A virus, human rhinoviruses, polio virus, retroviruses, rotaviruses, flaviviruses, hepaciviruses, togaviruses, and rubella virus.

8. The use of claim 1, wherein the vaccine comprises an inactivated orthomyxovirus.

9. The use of claim 8, wherein the vaccine comprises an inactivated influenza virus.

10. The use of claim 1, wherein the one or more attenuated viruses are selected from the group consisting of adenoviruses, picornaviruses, papillomaviruses, polyomaviruses, hepadnaviruses, parvoviruses, pox viruses, Epstein-Barr virus, cytomegalovirus (CMV), herpes viruses, roseolovirus, varicella zoster virus, filoviruses, paramyxoviruses, orthomyxoviruses, rhabdoviruses, arenaviruses, coronaviruses, human enteroviruses, hepatitis A virus, human rhinoviruses, polio virus, retroviruses, rotaviruses, flaviviruses, hepaciviruses, togaviruses, and rubella virus.

11. The use of claim 1, wherein the immunoprotective amount of a population of isolated homogenous allogeneic human mesenchymal stem cells is to be administered before the vaccine.

12. The use of claim 11, wherein the immunoprotective amount of a population of isolated homogenous allogeneic human mesenchymal stem cells is to be administered at least 1 week before the vaccine is administered.

13. The use of claim 11, wherein the immunoprotective amount of a population of isolated homogenous allogeneic human mesenchymal stem cells is to be administered at least 2 weeks before the vaccine is administered.

14. The use of claim 11, wherein the immunoprotective amount of a population of isolated homogenous allogeneic human mesenchymal stem cells is to be administered at least 3 weeks before the vaccine is administered.

15. The use of claim 11, wherein the immunoprotective amount of a population of isolated homogenous allogeneic human mesenchymal stem cells is to be administered at least 4 weeks before the vaccine is administered.

16. The use of claim 1, wherein the immunoprotective amount of a population of isolated homogenous allogeneic human mesenchymal stem cells is to be administered at least yearly for long-term enhancement of vaccine response.

17. The use of claim 1, wherein the immunoprotective amount of a population of isolated homogenous allogeneic human mesenchymal stem cells is a dose of about $20x10^6$ mesenchymal stem cells.

18. The use of claim 1, wherein the immunoprotective amount of a population of isolated homogenous allogeneic human mesenchymal stem cells is a dose of about $100x10^6$ mesenchymal stem cells.

19. The use of claim 1, wherein the immunoprotective amount of a population of isolated homogenous allogeneic human mesenchymal stem cells is a dose of about $200x10^6$ mesenchymal stem cells.

20. The use of claim 1, wherein the mesenchymal stem cells are obtained from a human donor and wherein a step of MHC matching of the human donor to the subject is not employed prior to the administration of the vaccine and mesenchymal stem cells to the subject.

21. The use of claim 1 wherein the vaccine and the immunoprotective amount of a population of isolated homogenous allogeneic human mesenchymal stem cells are to be repeatedly administered at least six months after the first administration of the vaccine.

22. The use of claim 1, wherein the intracellular TNF-α expression in B cells of the human subject decreases by at least two-fold as compared to the TNF-α expression levels in B cells of the human subject prior to administration of the adjuvant.

23. The use of claim 1, wherein the $CD4^+$:$CD8^+$ T cell ratio in the human subject increases by at least two-fold as compared to the $CD4^+$:$CD8^+$ T cell ratio in the human subject prior to administration of the adjuvant.

24. The use of claim 1, wherein the number of switched memory B cells in the human subject increases by at least two-fold as compared to the number of switched memory B cells in the human subject prior to administration of the adjuvant.

25. The use of claim 1, wherein the number of exhausted B cells in the human subject decreases by at least two-fold as compared to the number of exhausted B cells in the human subject prior to administration of the adjuvant.