CLAIMS

1. A method of manufacturing chimeric antigen receptor (CAR)-expressing T cells (CAR T cells) from peripheral blood mononuclear cells (PBMCs) from a subject from which a blood sample is obtained, comprising the steps of:

   a. obtaining PBMCs from the blood sample;

   b. isolating the PBMCs that have been obtained from the blood sample using a membrane filtration system and Ammonium-Chloride-Potassium (ACK) buffer;

   c. washing the PBMCs via centrifugation;

   d. washing the PBMCs using a membrane filtration system;

   e. manufacturing the CAR T cells from the PBMCs from step (d); and

   f. washing the CAR T cells from step (e) using a membrane filtration system.

2. The method of claim 1, wherein the method further comprises cryopreserving the PBMCs from step (c).

3. The method of claim 2, wherein the method further comprises thawing the cryopreserved PBMCs.

4. A method of manufacturing chimeric antigen receptor (CAR)-expressing T cells (CAR T cells) from peripheral blood mononuclear cells (PBMCs) from a leukapheresed blood sample from a subject, comprising the steps of:

   a. isolating the PBMCs from the leukapheresed blood sample using a membrane filtration system and Ammonium-Chloride-Potassium (ACK) buffer;

   b. cryopreserving the PBMCs from step (a);

   c. thawing the PBMCs from step (b);

   d. washing the thawed PBMCs from step (c) using a spinning membrane filtration system;

   e. manufacturing the CAR T cells from the PBMCs from step (d); and

   f. washing the CAR T cells from step (e) using a membrane filtration system.

5. A method of manufacturing chimeric antigen receptor (CAR)-expressing T cells (CAR T cells) from peripheral blood mononuclear cells (PBMCs) from a blood sample from a subject, comprising the steps of:

   a. isolating the PBMCs from the blood sample using a membrane filtration systemand Ammonium-Chloride-Potassium (ACK) buffer;

   b. cryopreserving and thawing the PBMCs from step (a);

   c. washing the thawed PBMCs from step (b) using a membrane filtration system;

   d. manufacturing the CAR T cells from the PBMCs; and

   e. washing of the CAR T cells using a spinning membrane filtration system.

6. The method of any one of claims 1 to 5, wherein any or all of the membrane filtration systems are a spinning membrane filtration system.

7. The method of claim 6, wherein the spinning membrane filtration system is a LOVO Automated Cell Processing System.

8. The method of any one of claims 1 to 7, wherein the ACK buffer comprises 50-300 mM ammonium chloride, 5-25 mM potassium carbonate, and 0.05-0.25 mM sodium EDTA.

9. The method of claim 8, wherein said ACK buffer comprises 150 mM ammonium chloride, 10 mM potassium carbonate, and 0.1 mM sodium EDTA.

10. The method of any one of claims 1 to 9, wherein said method improves reduction of platelets and red blood cells from said PBMCs by 18%-36% as compared to the same method using density gradient centrifugation in place of each use of membrane filtration.

11. The method of any one of claims 1 to 10, wherein said method improves recovery of CAR T cells after CAR T cell manufacturing by 17%-36% as compared to the same method using density gradient centrifugation in place of each use of membrane filtration.