CLAIMS

1. A method for lyophilising an exosome, the method comprising the steps of:

(a) providing an exosome suspension in a lyophilisation buffer comprising trehalose at a concentration of 4% w/v;

(b) freezing the exosome suspension; and

(c) removing water from the frozen suspension by freeze drying;

in which the lyophilised exosome exhibits at least one biological activity of an exosome.

2. A method according to Claim 1, in which:

(a) the exosome exhibits 5% or more, 10% or more, 15% or more, 20% or more, 25% or more, 30% or more, 35% or more, 40% or more, 45% or more, 50% or more, 55% or more, 60% or more, 65% or more, 70% or more, 75% or more, 80% or more, 85% or more, 90% or more, or 95% or more of the biological activity when reconstituted;

(b) the exosome exhibits the biological activity, such as therapeutic activity, when stored at a temperature of 0˚C or higher, 4˚C or higher, 10˚C or higher, 15˚C or higher, 20˚C or higher, 25˚C or higher, 30˚C or higher, 35˚C or higher, 40˚C or higher; or

(c) the exosome exhibits a biological activity when stored for 1 day or more, 5 days or more, 7 days or more, 10 days or more, 14 days or more, 20 days or more, 21 days or more, 25 days or more, 28 days or more, 30 days or more, 35 days or more, or 40 days or more;

as compared to an exosome which has not been lyophilised.

3. A method according to Claim 1 or 2, in which the biological activity comprises:

(a) CD73 enzymatic activity and in which the exosome exhibits 20% or more, such as 25% or more, 30% or more, 35% or more, 40% or more, 45% or more, 50% or more, 55% or more or 60% or more of CD73 enzymatic activity after 55 days of storage at 40˚C, preferably when compared to the activity after 1 day of storage at 40˚C;

(b) induction of polarization of CD4+ T cells to regulatory T cells (Tregs), in which the exosome exhibits 20% or more, such as 25% or more, 30% or more, 35% or more, 40% or more, 45% or more, 50% or more, 55% or more or 60% or more of activity to

induce polarization of CD4+ T cells to regulatory T cells (Tregs) after 3 weeks of storage at 40˚C, preferably when compared with a non-lyophilized exosome stored at -80°C; or

(c) cardioprotection, in which the exosome exhibits 20% or more, such as 25% or more, 30% or more, 35% or more, 40% or more, 45% or more, 50% or more, 55% or more or 60% or more of cardioprotective activity, preferably when compared with a non-lyophilized exosome stored at -80°C, preferably in which the cardioprotection is assayed using an *ex vivo* Landerdorf model of ischemia-reperfusion injury, preferably by the degree of hypercontracture over post-reperfusion time.

4. A method according to any preceding claim, in which the exosome comprises a mesenchymal stem cell (MSC) exosome.

5. A method according to any preceding claim, in which:

(i) step (b) comprises freezing the exosome suspension at a temperature of -10˚C or lower, -15˚C or lower or -20˚C or lower;

(ii) step (b) comprises freezing the exosome suspension for 1 hour or more, 2 hours or more or 3 hours or more;

(iii) step (c) comprises freeze drying at a temperature of -10˚C or lower, -15˚C or lower, -20˚C or lower, -25˚C or lower, -30˚C or lower, -35˚C or lower, -40˚C or lower or -45˚C or lower;

(iv) step (c) comprises freeze drying at a pressure of 1 mbar or lower, 0.5 mbar or lower, 0.25 mbar or lower, 0.1 mbar or lower or 0.05 mbar or lower;

(v) step (c) comprises freeze drying at a temperature of -10˚C or lower, -15˚C or lower, -20˚C or lower, -25˚C or lower, -30˚C or lower, -35˚C or lower, -40˚C or lower or -45˚C or lower; or

(vi) step (c) comprises freeze drying for 5 hours or longer, 10 hours or longer, 15 hours or longer or 20 hours or longer.

6. A method according to any preceding claim, in which step (b) comprises freezing the exosome suspension at -20˚C for 3 hours and/or step (c) comprises freeze drying at < 0.05mbar for 12 to 24 hours.

7. A method according to any preceding claim, in which the method further comprises a step before step (c) of transferring the frozen suspension to a freeze dryer pre-cooled to -48˚C.

8. A method according to any preceding claim, in which the method further comprises:

(a) storing the lyophilised exosome at a temperature of 4˚C or higher, such as 10˚C or higher, 15˚C or higher, 20˚C or higher, 25˚C or higher, 30˚C or higher, 35˚C or higher or 40˚C or higher;

(b) storing the lyophilised exosome in substantially dry conditions, such as in a desiccator, preferably in which the substantially dry conditions comprise storage in air with a relative humidity of 5% or less, such as 4% or less, 3% or less, 2% or less or 1% or less; or

(c) storing the lyophilised exosome in an inert gas, such as nitrogen.

9. A method according to any preceding claim, in which the lyophilisation buffer further comprises one or more of a buffer, for example a phosphate buffer, such as at pH 6.5-7.5, an amino acid for example L-glutamic acid, such as at 0.0049M and a protein binding stabiliser, for example human serum albumin such as at 1% w/v.

10. A method according to any preceding claim, in which the method further comprises a step of viral clearance, such as by exposure to gamma radiation, X-ray irradiation or ethylene oxide.

11. A method according to any preceding claim, comprising a step before step (b) of coating an object with the exosome suspension, such that removal of water from the frozen suspension deposits the exosome on the object, preferably in which the object comprises a scaffold made of a biocompatible material.

12. A method according to Claim 11, in which the object is selected from the group consisting of: a collagen membrane, a surgical dressing, a prosthetic bone replacement device, a cardiovascular device, a stent and a balloon catheter.