CLAIMS

We claim:

1. A method for purifying messenger RNA (mRNA), the method comprising the steps of:
    I. precipitating mRNA from a solution comprising one or more protein and/or short abortive transcript contaminants from manufacturing the mRNA to provide a suspension comprising precipitated mRNA;
    II. loading the suspension comprising the precipitated mRNA into a filtering centrifuge comprising a filter wherein the precipitated mRNA is retained by the filter;
    III. washing the retained precipitated mRNA by adding a wash buffer to the filtering centrifuge; and
    IV. recovering the retained precipitated mRNA from the filter;

wherein the filtering centrifuge is operated during loading step (b) and washing step (c) at a centrifuge speed that exerts a gravitational (g) force of less than 1300 g.

2. The method of claim 1, wherein centrifuge speed exerts a gravitational (g) force of between about 300 g and about 1300 g, between about 400 g and about 1100 g, between about 500 g and about 900 g, between about 700 g and about 900 g, between about 750 g and 850 g, between about 550 g and about 750 g, or between about 650 g and about 750.

3. The method of any one of the preceding claims, wherein the filtering centrifuge is operated at the same centrifuge speed during loading step (b) and washing step (c).

4. The method of any one of the preceding claims, wherein the recovering the retained precipitated mRNA from the filter comprises the steps of:
    i. solubilising the retained precipitated mRNA; and
    ii. collecting the solubilised mRNA.

5. The method of any one of the preceding claims, wherein step (a) further comprises adding at least one filtration aid to the suspension comprising precipitated mRNA.

6. The method of any one of the preceding claims, wherein the suspension comprises at least 100mg, 1g, 10g, 100g, 250g, 500g, 1kg, 10kg, or 100kg of mRNA.

7. The method of any one of the preceding claims, wherein the filter (i) comprises a porous substrate and/or a microfiltration membrane or ultrafiltration membrane.

8. The method of any one of the preceding claims, wherein the filter has an average pore size of about 0.5 micron or greater, about 0.75 micron or greater, about 1 micron or greater, about 2 microns or greater, about 3 microns or greater, about 4 microns or greater, or about 5 microns or greater, or about 0.01 micron to about 200 microns, about 1 micron to about 2000 microns, about 0.2 microns to about 5 microns, or about 1 micron to about 3 microns.

9. The method of any one of the preceding claims, wherein the volume of wash buffer for washing the retained precipitated mRNA is between about 0.5 L/g mRNA and about 8 L/g mRNA.

10. The method of claim 9, wherein the wash buffer is loaded into the filtering centrifuge at a rate of about 5 liter/min/m^2 to about 25 liter/min/m^2 (with respect to the surface area of the filter of the filtering centrifuge).

11. The method of any one of the proceeding claims, wherein the recovering the retained mRNA occurs (i) while the filtering centrifuge is in operation, or (ii) while the filtering centrifuge is not in operation.

12. The method of any one of claims 4-11, wherein the solubilising the retained mRNA comprises dissolving the mRNA in an aqueous medium.

13. The method of any one of claims 5-12, wherein the collecting of the solubilised mRNA comprises one or more steps of separating the filtration aid from the solubilised mRNA.

14. The method of any one of the preceding claims, wherein the filtering centrifuge is (i) a continuous centrifuge and/or the filtering centrifuge is orientated vertically or horizontally or the centrifuge is an inverted horizontal centrifuge, and/or (ii) comprises a sample feed port and/or a sample discharge port.

15. The method of any one of the preceding claims, wherein the mRNA suspension is loaded into the filtering centrifuge at a rate of about 1 liter/min to about 60 liter/min.

16. A purified mRNA obtained by the method of any one of claims 1-15.

17. A composition comprising the purified mRNA of claim 16.

18. The purified mRNA of claim 16 or the composition of claim 17 for use in therapy.

19. A process for purifying mRNA, the process comprising the steps of:

I. providing a suspension comprising precipitated mRNA in a first vessel, wherein the precipitated mRNA comprises one or more protein and/or short abortive transcript contaminants from manufacturing the mRNA;

II. providing a wash buffer in a second vessel;

III. transferring the content of the first vessel into a filtering centrifuge comprising a filter, wherein the transferring occurs at a rate of about 5 liter/min/m^2 to about 25 liter/min/m^2 (with respect to the surface area of the filter of the filtering centrifuge) while the filtering centrifuge is in operation at a first centrifuge speed such that the precipitated mRNA is retained on the filter of said filtering centrifuge;

IV. transferring the content of the second vessel into the filtering centrifuge, wherein the transferring occurs at a rate of about 5 liter/min/m^2 to about 25 liter/min/m^2 (with respect to the surface area of the filter of the filtering centrifuge)while the filtering centrifuge remains in operation at the first centrifuge speed, thereby washing the precipitated mRNA retained on the filter of said filtering centrifuge with the wash buffer; and

V. recovering the washed precipitated mRNA from the filter of said filtering centrifuge.

20. A composition comprising mRNA, amphiphilic polymer and a filtration aid at relative concentrations of about 1:1:10 in a sterile, RNase-free container.