CLAIMS

1. A method of assessing the potency of a sample batch of defibrotide comprising:

growing mammalian cells in culture;

incubating the cells with a solution containing at least one cytotoxic agent and at least one concentration of defibrotide from the sample batch;

measuring the viability of the cells after an incubation period;

comparing the cell viability to the cell viability for a reference batch of defibrotide; and

calculating the potency of the sample batch of defibrotide based on the comparison.

2. The method of claim 1, wherein the cells are incubated with the cytotoxic agent and at least four different concentrations of defibrotide from the sample batch, and wherein the cell viability is determined for each of the concentrations to create a dose-response curve.

3. The method of claim 1, wherein the cell viability measured for the sample batch of defibrotide is compared to a calibration curve obtained from cell viability measurements with the reference batch of defibrotide.

4. The method of claim 1, wherein calculating the potency of the defibrotide sample batch comprises determining a potency ratio relative to the potency of a reference batch of defibrotide.

5. The method of claim 1, wherein the mammalian cells are human endothelial cells, human epithelial cells, human liver sinusoidal endothelial cells, or human microvascular endothelial cells.

6. The method of claim 1, wherein the mammalian cells are present at a density of about 5×10^4 cells/ml to about 5×10^5 cells/ml.

7. The method of claim 1, wherein the cytotoxic agent is fludarabine, 9-beta-D-arabinofuranosyl-2-fluoroadenine (F-Ara-A), or doxorubicin.

8. The method of claim 7, wherein fludarabine or F-Ara-A is present in the solution at a concentration of about 10 μg/ml to about 50 μg/ml or wherein doxorubicin is present in the solution at a concentration of about 0.1 μg/ml to about 10 μg/ml.

9. The method of claim 1, wherein the at least one concentration of defibrotide from the sample batch is in the range of about 1 μg/ml to about 100 μg/ml.

10. The method of claim 1, wherein the incubation period is at least 24 hours, at least 48 hours, or at least 72 hours.

11. The method of claim 1, wherein measuring cell viability comprises performing a colorimetric assay based on the reduction of tetrazolium dyes.

12. The method of claim 11, wherein the tetrazolium dye is 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide or 2-(2-methoxy-4-nitrophenyl)-3-(4-nitrophenyl)-5-(2,4-disulfophenyl)-2H-tetrazolium.

13. The method of claim 1, wherein measuring cell viability comprises measuring the absorbance of the solution following incubation with the cells.

14. The method of claim 1, wherein the sample batch of defibrotide is extracted from bovine tissue or porcine tissue.

15. A method of assessing the potency of a sample batch of defibrotide comprising:

growing mammalian cells in culture in a multi-well microtiter plate;

incubating the cells with a solution containing at least one cytotoxic agent and at least four different concentrations of defibrotide from the sample batch;

collecting dose response data points simultaneously;

measuring the viability of the cells after an incubation period;

comparing the cell viability to the cell viability for a reference batch of defibrotide; and

calculating the potency of the sample batch of defibrotide based on the comparison.

16. The method of claim 15, wherein the cells are incubated with a solution containing defibrotide at a concentration between 1.25 to 80 µg/mL.

17. The method of claim 15, wherein the cell viability is determined for each of the concentrations to create a dose-response curve.

18. The method of claim 15, wherein the cytotoxic agent fludarabine or F-Ara-A is present in the solution at a concentration of about 10 µg/ml.

19. The method of claim 15, wherein the multi-well microtiter plate is a 96-well plate.

20. The method of claim 15, wherein at least 3 replicates of incubating the cells with a solution containing at least one cytotoxic agent and at least four different concentrations of defibrotide from the sample batch is performed.