

CLAIMS

1. Use of a lipid nano particle (LNP) composition comprising an effective amount of:
 - i. an mRNA encoding a Cas nuclease, and
 - ii. a guide RNA that targets the TTR genein the manufacture of a medicament for the treatment of amyloidosis associated with TTR (ATTR) in a human subject,
wherein the medicament is adapted to reduce serum TTR relative to baseline serum TTR on systemic administration.
2. Use of a lipid nano particle (LNP) composition comprising:
 - i. an mRNA encoding a Cas nuclease, and
 - ii. a guide RNA that targets the TTR genein the manufacture of a medicament for editing the transthyretin (TTR) gene in a human subject having amyloidosis associated with TTR (ATTR),
wherein the medicament is adapted for editing the gene at the site targeted by the guide RNA in a hepatocyte of the subject; and
wherein the medicament is adapted to change a level of a biosafety metric in the subject on systemic administration to a level that is acceptable as compared to a baseline level of the biosafety metric.
3. Use of a LNP composition comprising an effective amount of:
 - i. an mRNA encoding a Cas nuclease, and
 - ii. a guide RNA that targets the TTR genein the manufacture of a medicament for editing the transthyretin (TTR) gene in a human subject having amyloidosis associated with TTR (ATTR),
wherein the medicament is adapted for editing the TTR gene at the site targeted by the guide RNA in a hepatocyte of the subject; and
wherein the medicament is adapted to produce a clinically significant improvement in a level of a clinical metric in the subject on systemic administration as compared to a baseline level.
4. Use of a LNP composition comprising an effective amount of:
 - i. an mRNA encoding a Cas nuclease, and
 - ii. a guide RNA that targets the TTR genein the manufacture of a medicament for treating amyloidosis associated with TTR (ATTR) in a human subject, wherein the medicament is adapted to change a level of a biosafety metric in the subject on systemic administration to a level that is acceptable as compared to a baseline level.
5. Use of a LNP composition comprising an effective amount of:
 - i. an mRNA encoding a Cas nuclease, and
 - ii. a guide RNA that targets the TTR genein the manufacture of a medicament for treating amyloidosis associated with TTR (ATTR) in a human subject, wherein the medicament is adapted to produce a clinically significant improvement in a level of a clinical metric in the subject on systemic administration as compared to a baseline level of the clinical metric.

6. Use of a LNP composition comprising an effective amount of:
- i. an mRNA encoding a Cas nuclease, and
 - ii. a guide RNA that targets the TTR gene
- in the manufacture of a medicament for treating amyloidosis associated with TTR (ATTR) in a human subject,
wherein the medicament is adapted for systemic administration and wherein the medicament is adapted to administer the mRNA encoding a Cas nuclease and the guide RNA that targets the TTR gene at a combined dose of about 25 to about 100 mg.
7. Use of a LNP composition comprising:
- i. an mRNA encoding a Cas nuclease, and
 - ii. a guide RNA that targets a gene in the liver
- in the manufacture of a medicament for in vivo editing of a gene in the liver of a human subject having a monogenic disorder,
wherein the medicament is adapted for editing the gene at the site targeted by the guide RNA in a hepatocyte of the subject; and
wherein the medicament is adapted to change a level of a biosafety metric in the subject on systemic administration to a level that is acceptable as compared to a baseline level of the biosafety metric.
8. Use of a LNP composition comprising an effective amount of:
- i. an mRNA encoding a Cas nuclease, and
 - ii. a guide RNA that targets a gene in the liver
- in the manufacture of a medicament for treating a human subject having a monogenic disorder,
wherein the medicament is adapted for editing the gene in the liver thereby treating the monogenic disorder, and wherein the treatment is safe and well-tolerated for systemic administration.
9. Use of a LNP composition comprising an effective amount of:
- i. an mRNA encoding a Cas nuclease, and
 - ii. a guide RNA that targets a gene in the liver
- in the manufacture of a medicament for treating a human subject having a monogenic disorder,
wherein the subject has a first level of a biosafety metric prior to systemic administration of the medicament and a second level of the biosafety metric in a period of time after systemic administration of the medicament, and
wherein the medicament is adapted to change a level of the biosafety metric in the subject on systemic administration to a level that is acceptable as compared to a baseline level.
10. The use of any one of claims 1-9, wherein the human subject has amyloidosis associated with TTR (ATTR) prior to the systemic administration.
11. The use of any one of claims 7-9, wherein the human subject has a monogenic disorder prior to the systemic administration.

12. The use of any one of claims 1-11, wherein the LNP comprises (9Z, 12Z)-3-((4,4-bis(octyloxy)butanoyl)oxy)-2-((((3-(diethylamino)propoxy)carbonyl)oxy)methyl)propyl octadeca-9, 12-dienoate.
13. The use of any one of claims 1-12, wherein the LNP comprises a PEG lipid.
14. The use of claim 13, wherein the PEG lipid comprises dimyristoylglycerol (DMG), e.g., PEG-2k.
15. The use of any one of claims 1-14, wherein the LNP composition has an N/P ratio of about 5-7.
16. The use of any one of claims 1-15, wherein the guide RNA and the mRNA encoding the Cas nuclease are present in a ratio ranging from about 5:1 to about 1:5 by weight.
17. The use of any one of claims 1-16, wherein the mRNA encodes a Class 2 Cas nuclease, e.g., a Cas9 nuclease such as a *S. pyogenes* Cas9 nuclease.
18. The use of any one of claims 1-17, wherein the mRNA encoding the Cas nuclease is codon-optimized.
19. The use of any one of claims 1-18, wherein at least one of the guide RNA and the mRNA comprise a modification.
20. The use of claim 19, wherein the modification includes a 2'-O-methyl modified nucleotide or a phosphorothioate bond between nucleotides.
21. The use of any one of claims 7-9, wherein the monogenic disorder is ATTR.
22. The use of any one of claims 7-9, wherein the gene in the liver is *TTR*.
23. The use of any one of claims 1-6, wherein the ATTR is hereditary transthyretin amyloidosis.
24. The use of any one of claims 1-6, wherein the ATTR is wild-type transthyretin amyloidosis.
25. The use of any one of claims 1-6, wherein the ATTR is hereditary transthyretin amyloidosis with polyneuropathy.
26. The use of any one of claims 1-6 or 23 wherein the ATTR is hereditary transthyretin amyloidosis with cardiomyopathy.
27. The use of claim 24, wherein the ATTR is wildtype transthyretin amyloidosis with cardiomyopathy.
28. The use of claim 26 or 27, wherein the subject is classified under the New York Health Association (NYHA) classification as Class I, Class II, or Class III.

29. The use of any one of claims 2, 4, 7, or 9, wherein the medicament is adapted to change a level of the biosafety metric in the subject to a level that is acceptable as compared to a baseline level of the biosafety metric.
30. The use of any one of claims 2, 4, 7, or 9, wherein the biosafety metric is at least one of prothrombin, activated partial thromboplastin time (aPTT), fibrinogen, alanine aminotransferase (ALT), and aspartate aminotransferase (AST).
31. The use of claim 3 or 5, wherein the clinical metric is serum TTR level.
32. The use of any one of claims 1-31, wherein the medicament is adapted to administer the mRNA encoding a Cas nuclease and the guide RNA that targets the TTR gene at a combined dose of about 0.3 mg/kg.
33. The use of any one of claims 1-31, wherein the medicament is adapted to administer the mRNA encoding a Cas nuclease and the guide RNA that targets the TTR gene at a combined dose of about 0.7 mg/kg.
34. The use of any one of claims 1-31, wherein the medicament is adapted to administer the mRNA encoding a Cas nuclease and the guide RNA that targets the TTR gene at a combined dose of about 1.0 mg/kg.
35. The use of any one of claims 1-31, wherein the medicament is adapted to administer the mRNA encoding a Cas nuclease and the guide RNA that targets the TTR gene at a combined dose of about 50 mg of total RNA.
36. The use of any one of claims 1-31, wherein the medicament is adapted to administer the mRNA encoding a Cas nuclease and the guide RNA that targets the TTR gene at a combined dose of about 55 mg of total RNA.
37. The use of any one of claims 1-31, wherein the medicament is adapted to administer the mRNA encoding a Cas nuclease and the guide RNA that targets the TTR gene at a combined dose of about 80 mg of total RNA.
38. The use of claim 37, wherein the medicament is adapted to administer a second dose of the LNP composition, wherein administration of the second dose reduces serum TTR level by at least 80% relative to the baseline serum TTR level.
39. The use of claim 37, wherein the medicament is adapted to administer a second dose of the LNP composition, wherein administration of the second dose reduces serum TTR level by at least 80% relative to the baseline serum TTR level (i) prior to administration of the first dose or (2) prior to administration of the second dose and after administration of the first dose.
40. The use of any one of claims 35-39, wherein the total RNA about is within $\pm 5\%$ of the combined mg dose.
41. The use of any one of claims 35-39, wherein the total RNA about is within $\pm 10\%$ of the combined mg dose.

42. The use of any one of claims 1-41, wherein the medicament is adapted to reduce or knock down expression of the TTR gene on administration.
43. The use of any one of claims 1-42, wherein the medicament is adapted to reduce or knock down expression of the TTR gene by at least 60%, 70%, 80%, 85%, 90%, 95%, 96%, 98%, 99% or by 100%, as compared to baseline before administration of the composition.
44. The use of any one of claims 1-43, the medicament is adapted to reduce serum TTR level in the subject by at least 60%, 70%, 80%, 85%, 90%, 95%, 96%, 98%, 99% or by 100% as compared to serum TTR level before administration of the composition.
45. The use of any one of claims 1-43, wherein the medicament is adapted to reduce serum TTR levels to less than about 50 $\mu\text{g/mL}$, 40 $\mu\text{g/mL}$, 30 $\mu\text{g/mL}$, 20 $\mu\text{g/mL}$, or 10 $\mu\text{g/mL}$ after administration of the composition.
46. The use of any one of claims 1-45, wherein the medicament is adapted to administer with a second therapeutic.
47. The use of claim 46, wherein the second therapeutic is diflunisal or tafamidis.