METHOD FOR CONDUCTING SOLID STATE NMR ON MACROMOLECULE-CONTAINING SOLID STATE FORMULATIONS

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims the benefit of priority of U.S. Provisional patent application no. 62/909,918, filed October 3, 2019, the disclosure of which is hereby incorporated by reference in its entirety.

BACKGROUND

[0002] The present disclosure relates to methods of conducting solid state nuclear magnetic resonance ("ssNMR") spectroscopy on macromolecule-containing solid state formulations, such as antibody-containing pharmaceutical formulations that have been lyophilized or frozen, and using the methods disclosed herein to determine, for example, the stability of the formulation, the degree of aggregation in the formulation, and/or the degree of molecular mobility in the formulation.

[0003] Pharmaceutical formulations are often prepared in the solid state, such as frozen or lyophilized, to help preserve the integrity of the active pharmaceutical ingredient ("API") in the formulation during storage. Maintaining the stability of the API in the solid state is key for ensuring formulation quality. Current methods for understanding the stability of such a solid state formulation typically involve storing the solid state (e.g., lyophilized or frozen) formulation for a period of time, reconstituting or thawing the formulation to its liquid state, and then determining formulation stability using, for example, size exclusion chromatography ("SEC"). These liquid state stability studies are often time-consuming, resulting in delayed drug development and/or shorter-than-desired shelf life. Thus, there is a need to accurately determine the stability of a solid state formulation, such as a lyophilized or frozen pharmaceutical formulation, in real time to improve the time required for formulation development, and to reduce the risk of reformulation after years of development.

SUMMARY

[0004] In one aspect, the present disclosure provides a method of conducting direct detection ^{1}H solid state NMR ("ssNMR") on a macromolecule-containing solid state formulation, the method comprising: (a) equilibrating a solid state formulation comprising a macromolecule at a first temperature; (b) conducting a ^{1}H spin-lattice relaxation time ("T_1") experiment on the solid state formulation at the first temperature using direct detection ^{1}H ssNMR to generate at least

three free induction decay (“FID”) plots at the first temperature, wherein the T_1 experiment comprises baseline suppression and a saturation recovery sequence having at least three variable delay times from which each FID plot is generated; (c) equilibrating the solid state formulation at a second temperature, and repeating step (b) at the second temperature to generate at least three FID plots at the second temperature; (d) equilibrating the solid state formulation at a third temperature, and repeating step (b) at the third temperature to generate at least three FID plots at the third temperature; (e) generating a saturation recovery curve at each temperature; and (f) fitting each saturation recovery curve to a nonlinear regression equation to generate a T_1 value at each temperature. In one example, the disclosure provides a method of conducting direct detection ^{1}H solid state NMR (“ssNMR”) on a macromolecule-containing solid state formulation, the method comprising: (a) equilibrating a solid state formulation comprising a macromolecule at a first temperature; (b) conducting a ^{1}H spin-lattice relaxation time (“T_1”) experiment on the solid state formulation at the first temperature using direct detection ^{1}H ssNMR to generate at least three free induction decay (“FID”) plots at the first temperature, wherein the T_1 experiment comprises

a saturation recovery sequence having at least three variable delay times from which each FID plot is generated; (c) equilibrating the solid state formulation at a second temperature and repeating step (b) at the second temperature to generate at least three FID plots at the second temperature; (d) equilibrating the solid state formulation at a third temperature and repeating step (b) at the third temperature to generate at least three FID plots at the third temperature; (e) generating a saturation recovery curve at each temperature; and (f) fitting each saturation recovery curve to a nonlinear regression equation to generate a T_1 value at each temperature. In some cases, the method further comprises: (g) converting each T_1 value to ^1H spin-lattice relaxation rate ("R_1"), and (h) plotting R_1 versus temperature to generate a relaxation rate curve for the solid state formulation. In some examples, the method further comprises analyzing the relaxation rate curve to determine the molecular mobility of the macromolecule in the solid state formulation, the degree of aggregation in the solid state formulation, and/or the stability of the solid state formulation. In various examples, the T_1 experiment comprises baseline suppression. In various cases, the T_1 experiment comprises magic angle spinning. In some cases, the method excludes one or both of retuning and recalibrating the ssNMR probe after equilibrating at the first temperature.

[0005] In some cases, step (d) is repeated at 5 or more additional temperatures. In some examples, step (d) is repeated at 10 or more additional temperatures. In various cases step (d) is repeated at 25 or more additional temperatures.

[0006] In some examples, each temperature is in a range from about -200 °C to about 150 °C. In various examples, each temperature is in a range from about -100 °C to about 150 °C. In some cases, each temperature is in a range from about -50 °C to about 150 °C. In various cases, the highest and lowest temperatures have a difference of at least about 25 °C. In some examples, the lowest temperature is in a range from about -200 °C to about 25 °C. In various examples, the lowest temperature is in a range from about -100 °C to about 0 °C. In some cases, the lowest temperature is in a range from about -50 °C to about -30 °C.

[0007] In various cases, in each equilibrating step the solid state formulation is held at the temperature for a duration in the range of about one minute to about one hour before conducting the T_1 experiment. In some examples, in each equilibrating step the solid state formulation is held at the temperature for a duration in the range of about one minute to about 10 minutes before conducting the T_1 experiment. In various examples, the solid state formulation is held at the temperature for about five minutes before conducting the T_1 experiment.

[0008] In some cases, each T_1 experiment comprises at least five variable delay times at each temperature to generate at least five FID plots at each temperature. In some examples, each T_1 experiment comprises at least six variable delay times at each temperature to generate at least six FID plots at each temperature. In various cases, each T_1 experiment comprises at least eight variable delay times at each temperature to generate at least eight FID plots at each temperature. In some examples, each T_1 experiment comprises at least nine variable delay times at each temperature to generate at least nine FID plots at each temperature.

[0009] In some examples, the saturation recovery curve is generated by: (a) plotting the signal intensity of each of the at least three FID plots versus delay time; or (b) Fourier transforming each of the at least three FID plots to generate a plot of intensity versus frequency; and (i) plotting peak height versus delay time; or (ii) plotting integral peak intensity versus delay time. In various examples, the saturated recovery curve is generated by plotting the signal intensity of the FID plot versus delay time.

[0010] In some cases, the T_1 experiment comprises a magnetic field having a frequency in a range from about 200 MHz to about 1.2 GHz. In various cases, the frequency range is from about 300 MHz to about 1 GHz. In various examples, the frequency is about 500 MHz.

[0011] In some examples, all of the T_1 experiments are conducted within a time period of up to 48 hours. In various examples, all of the T_1 experiments are conducted within a time period of up to 24 hours. In some cases, all of the T_1 experiments are conducted within a time period of up to 12 hours. In various cases, all of the T_1 experiments are conducted within a time period of up to 6 hours.

[0012] In some examples, the macromolecule is a biologic molecule. In various examples, the biologic molecule is a protein. In some cases, the protein is an antibody or a bispecific antibody construct.

[0013] In various cases, the solid state formulation is a lyophilized formulation. In some examples, the fitting of step (f) is monoexponential. In various examples, the highest temperature is in a range from about 50 °C to about 150 °C. In some cases, the highest temperature is in a range from about 100 °C to about 150 °C. In various cases, the highest and lowest temperatures have a difference of at least about 50 °C. In some examples, the T_1 experiment comprises a variable delay period in a range from about 0.01 seconds to about 60 seconds.

[0014] In some cases, the solid state formulation is a frozen formulation. In various cases, the fitting of step (f) is biexponential. In some examples, the highest temperature is from about -15 °C to about 0 °C. In some examples, the highest temperature is from about -15 °C to about -10 °C. In some cases, the highest and lowest temperatures have a difference of at least about 40 °C. In some examples, each T_1 experiment comprises nine variable delay times at each temperature to generate nine FID plots at each temperature. In various cases, T_1 experiment comprises a variable delay period in a range from about 0.1 seconds to about 240 seconds.

[0015] The method of claim 1, wherein the solid state formulation is a lyophilized formulation and: the saturation recovery curve is generated by plotting the signal intensity of each of the at least three FID plots versus delay time; the T_1 experiment comprises baseline suppression and magic angle spinning; steps (d)-(f) are repeated at 15 or more temperatures; each temperature is in a range from about -50 °C to about 150 °C; the highest and lowest temperatures have a difference from about 75 °C to about 100 °C; in each equilibrating step the solid state formulation is held at the temperature for a duration in the range of about one minute to about ten minutes before conducting the T_1 experiment; the method excludes retuning and recalibrating the ssNMR probe after equilibrating at the first temperature; the variable delay period is in a range from about 0.1 seconds to about 60 seconds; and each T1 experiment comprises six variable delay times at each temperature to generate six FID plots at each temperature. In some examples, the method further comprises: (g) converting each T_1 value to ^{1}H spin-lattice relaxation rate ("R_1"), and (h) plotting R_1 versus temperature to generate a relaxation rate curve for the solid state formulation. In various examples, the method further comprises analyzing the relaxation rate curve to determine the molecular mobility of the macromolecule in the solid state formulation, the degree of aggregation in the solid state formulation, and/or the stability of the solid state formulation. In some cases, the macromolecule is a biologic molecule. In various cases, the biologic molecule is a protein.

[0016] The method of claim 1, wherein the solid state formulation is a frozen formulation and: the saturation recovery curve is generated by plotting the signal intensity of each of the at least three FID plots versus delay time; the T_1 experiment comprises baseline suppression and magic angle spinning; steps (d)-(f) are repeated at 25 or more temperatures; each temperature is in a range from about -50 °C to about 0 °C; the highest and lowest temperatures have a difference from about 25 °C to about 40 °C; in each equilibrating step the solid state formulation is held at the temperature for a duration in the range of about one minute to about ten minutes before conducting the T_1 experiment; the method excludes retuning and recalibrating the ssNMR probe

after equilibrating at the first temperature; the variable delay period is in a range from about 0.01 seconds to about 240 seconds; and each T_1 experiment comprises nine variable delay times at each temperature to generate nine FID plots at each temperature. In some examples, the method further comprises: (g) converting each T_1 value to ^{1}H spin-lattice relaxation rate ("R_1"), and (h) plotting R_1 versus temperature to generate a relaxation rate curve for the solid state formulation. In various examples, the method further comprises analyzing the relaxation rate curve to determine the molecular mobility of the macromolecule in the solid state formulation, the degree of aggregation in the solid state formulation, and/or the stability of the solid state formulation. In some cases, the macromolecule is a biologic molecule. In various cases, the biologic molecule is a protein.

[0017] In another aspect, the present disclosure provides a method of selecting a macromolecule-containing solid state formulation among a group of test macromolecule-containing solid state formulations, the method comprising: (I) generating a relaxation rate curve for each macromolecule-containing solid state formulation in the group of test macromolecule-containing solid state formulations; wherein the relaxation rate curve for each macromolecule-containing solid state formulation is generated by: (a) equilibrating the solid state formulation at a first temperature; (b) conducting a ^{1}H spin-lattice relaxation time ("T_1") experiment on the solid state formulation at the first temperature using direct detection ^{1}H ssNMR to generate at least three free induction decay ("FID") plots at the first temperature, wherein the T_1 experiment comprises baseline suppression and a saturation recovery sequence having at least three variable delay times from which each FID plot is generated; (c) equilibrating the solid state formulation at a second temperature, and repeating step (b) at the second temperature to generate at least three FID plots at the second temperature; (d) equilibrating the solid state formulation at a third temperature and repeating step (b) at the third temperature to generate at least three FID plots at the third temperature; (e) generating a saturation recovery curve at each temperature; (f) fitting each saturation recovery curve to a nonlinear regression equation to generate a T_1 value at each temperature; (g) converting each T_1 value to ^{1}H spin-lattice relaxation rate ("R_1"), and (h) plotting R_1 versus temperature to generate a relaxation rate curve for the solid state formulation; (II) comparing the maximum R_1 peak value of each curve, the temperature of the maximum R_1 peak of each curve, the width of the maximum R_1 peak of each curve, or a combination thereof; and (III) selecting the solid state formulation which has the lowest maximum R_1 peak value, the highest temperature of the maximum R_1 peak, or the narrowest R_1 peak width.

Another example of the disclosure provides a method of selecting a macromolecule-containing solid state formulation among a group of test macromolecule-containing solid state formulations, the method comprising: (I) generating a relaxation rate curve for each macromolecule-containing solid state formulation in the group of test macromolecule-containing solid state formulations; wherein the relaxation rate curve for each macromolecule-containing solid state formulation is generated by: (a) equilibrating the solid state formulation at a first temperature; (b) conducting a ^{1}H spin-lattice relaxation time ("T_1") experiment on the solid state formulation at the first temperature using direct detection ^{1}H ssNMR to generate at least three free induction decay ("FID") plots at the first temperature, wherein the T_1 experiment comprises a saturation recovery sequence having at least three variable delay times from which each FID plot is generated; (c) equilibrating the solid state formulation at a second temperature, and repeating step (b) at the second temperature to generate at least three FID plots at the second temperature; (d) equilibrating the solid state formulation at a third temperature and repeating step (b) at the third temperature to generate at least three FID plots at the third temperature; (e) generating a saturation recovery curve at each temperature; (f) fitting each saturation recovery curve to a nonlinear regression equation to generate a T_1 value at each temperature; (g) converting each T_1 value to ^{1}H spin-lattice relaxation rate ("R_1"), and (h) plotting R_1 versus temperature to generate a relaxation rate curve for the solid state formulation; (II) comparing the maximum R_1 peak value of each curve, the temperature of the maximum R_1 peak of each curve, the width of the maximum R_1 peak of each curve, or a combination thereof; and (III) selecting the solid state formulation which has the lowest maximum R_1 peak value, the highest temperature of the maximum R_1 peak, or the narrowest R_1 peak width.

[0018] In yet another aspect, the present disclosure provides a method of selecting a formulation excipient for use in a macromolecule-containing solid state formulation, the method comprising: (I) generating a relaxation rate curve for each macromolecule-containing solid state formulation in a group of test macromolecule-containing solid state formulations, each formulation having a different composition of excipients, a different amount of one or more excipients, or both; wherein the relaxation rate curve for each macromolecule-containing solid state formulation is generated by: (a) equilibrating the solid state formulation at a first temperature; (b) conducting a ^{1}H spin-lattice relaxation time ("T_1") experiment on the solid state formulation at the first temperature using direct detection ^{1}H ssNMR to generate at least three free induction decay ("FID") plots at the first temperature, wherein the T_1 experiment comprises baseline suppression and a saturation recovery sequence having at least three variable delay times from which each FID plot is generated;

(c) equilibrating the solid state formulation at a second temperature, and repeating step (b) at the second temperature to generate at least three FID plots at the second temperature; (d) equilibrating the solid state formulation at a third temperature, and repeating step (b) at the third temperature to generate at least three FID plots at the third temperature; (e) generating a saturation recovery curve at each temperature; (f) fitting each saturation recovery curve to a nonlinear regression equation to generate a T_1 value at each temperature; (g) converting each T_1 value to ^1H spin-lattice relaxation rate ("R_1"), and (h) plotting R_1 versus temperature to generate a relaxation rate curve for the solid state formulation; (II) comparing the maximum R_1 peak value of each curve, the temperature of the maximum R_1 peak of each curve, the width of the maximum R_1 peak of each curve, or a combination thereof; and (III) selecting an excipient that is present in the solid state formulation that has the lowest maximum R_1 peak value, the highest temperature of the maximum R_1 peak, or the narrowest R_1 peak width. Yet another example of the disclosure provides a method of selecting a formulation excipient for use in a macromolecule-containing solid state formulation, the method comprising:

(I) generating a relaxation rate curve for each macromolecule-containing solid state formulation in a group of test macromolecule-containing solid state formulations, each formulation having a different composition of excipients, a different amount of one or more excipients, or both; wherein the relaxation rate curve for each macromolecule-containing solid state formulation is generated by: (a) equilibrating the solid state formulation at a first temperature; (b) conducting a ^{1}H spin-lattice relaxation time ("T_1") experiment on the solid state formulation at the first temperature using direct detection ^{1}H ssNMR to generate at least three free induction decay ("FID") plots at the first temperature, wherein the T_1 experiment comprises a saturation recovery sequence having at least three variable delay times from which each FID plot is generated; (c) equilibrating the solid state formulation at a second temperature, and repeating step (b) at the second temperature to generate at least three FID plots at the second temperature; (d) equilibrating the solid state formulation at a third temperature, and repeating step (b) at the third temperature to generate at least three FID plots at the third temperature; (e) generating a saturation recovery curve at each temperature; (f) fitting each saturation recovery curve to a nonlinear regression equation to generate a T_1 value at each temperature; (g) converting each T_1 value to ^{1}H spin-lattice relaxation rate ("R_1"), and (h) plotting R_1 versus temperature to generate a relaxation rate curve for the solid state formulation; (II) comparing the maximum R_1 peak value of each curve, the temperature of the maximum R_1 peak of each curve, the width of the maximum R_1 peak of each curve, or a combination thereof; and (III) selecting an excipient that is present in the solid state formulation that has the lowest maximum R_1 peak value, the highest temperature of the maximum R_1 peak, or the narrowest R_1 peak width.

[0019] Further examples and advantages will be apparent to those of ordinary skill in the art from a review of the following detailed description. While the methods disclosed herein are susceptible of examples in various forms, the description hereafter includes specific examples with the understanding that the disclosure is illustrative, and is not intended to limit the disclosure to the specific examples described herein.

BRIEF DESCRIPTION OF THE DRAWINGS

[0020] FIG. 1 shows the pulse sequence for the ssNMR methods of the disclosure including the baseline suppression sequence.

[0021] FIG. 2 shows a comparison of ^{1}H R_1 relaxation rates measured using a saturation recovery sequence combined with various detection methods for lyophilized 25:1 trehalose: anti-streptavidin. "BLS" stands for baseline suppression.

WHAT IS CLAIMED:

1. A method of conducting direct detection ^{1}H solid state NMR ("ssNMR") on a macromolecule-containing solid state formulation, the method comprising:

(a) equilibrating a solid state formulation comprising a macromolecule at a first temperature;

(b) conducting a ^{1}H spin-lattice relaxation time ("T_1") experiment on the solid state formulation at the first temperature using direct detection ^{1}H ssNMR to generate at least three free induction decay ("FID") plots at the first temperature, wherein the T_1 experiment comprises baseline suppression and a saturation recovery sequence having at least three variable delay times from which each FID plot is generated;

(c) equilibrating the solid state formulation at a second temperature, and repeating step (b) at the second temperature to generate at least three FID plots at the second temperature;

(d) equilibrating the solid state formulation at a third temperature, and repeating step (b) at the third temperature to generate at least three FID plots at the third temperature;

(e) generating a saturation recovery curve at each temperature; and

(f) fitting each saturation recovery curve to a nonlinear regression equation to generate a T_1 value at each temperature.

2. The method of claim 1, further comprising:

(g) converting each T_1 value to ^{1}H spin-lattice relaxation rate ("R_1"), and

(h) plotting R_1 versus temperature to generate a relaxation rate curve for the solid state formulation.

3. The method of claim 2, further comprising analyzing the relaxation rate curve to determine the molecular mobility of the macromolecule in the solid state formulation, the degree of aggregation in the solid state formulation, and/or the stability of the solid state formulation.

4. The method of any one of claims 1-3, further comprising repeating step (d) at 5 or more additional temperatures.

5. The method of claim 4, wherein step (d) is repeated at 10 or more additional temperatures.

6. The method of claim 5, wherein step (d) is repeated at 25 or more additional temperatures.

7. The method of any one of claims 1-6, wherein each temperature is in a range from -200 °C to 150 °C.

8. The method of claim 7, wherein each temperature is in a range from -100 °C to 150 °C.

9. The method of claim 8, wherein each temperature is in a range from -50 °C to 150 °C.

10. The method of any one of claims 1-9, wherein the highest and lowest temperatures have a difference of at least 25 °C.

11. The method of any one of claims 1-10, wherein the lowest temperature is in a range from -200 °C to 25 °C.

12. The method of claim 11, wherein the lowest temperature is in a range from -100 °C to 0 °C.

13. The method of claim 12 wherein the lowest temperature is in a range from -50 °C to -30 °C.

14. The method of any one of claims 1-13, wherein the T_1 experiment comprises baseline suppression.

15. The method of any one of claim 1-14, wherein in each equilibrating step the solid state formulation is held at the temperature for a duration in the range of one minute to one hour before conducting the T_1 experiment.

16. The method of claim 15, wherein in each equilibrating step the solid state formulation is held at the temperature for a duration in the range of one minute to 10 minutes before conducting the T_1 experiment.

17. The method of claim 16, wherein the solid state formulation is held at the temperature for about five minutes before conducting the T_1 experiment.

18. The method of any one of claims 1-17, wherein said method excludes one or both of retuning and recalibrating the ssNMR probe after equilibrating at the first temperature.

19. The method of any one of claims 1-18, wherein each T_1 experiment comprises at least five variable delay times at each temperature to generate at least five FID plots at each temperature.

20. The method of claim 19, wherein each T_1 experiment comprises nine variable delay times at each temperature to generate nine FID plots at each temperature.

21. The method of any one of claims claim 1-20, wherein the saturation recovery curve is generated by:

(a) plotting the signal intensity of each of the at least three FID plots versus delay time; or

(b) Fourier transforming each of the at least three FID plots to generate a plot of intensity versus frequency; and

(i) plotting peak height versus delay time; or

(ii) plotting integral peak intensity versus delay time.

22. The method of claim 21, wherein the saturated recovery curve is generated by plotting the signal intensity of the FID plot versus delay time.

23. The method of any one of claims 1-22, wherein the T_1 experiment comprises a magnetic field having a frequency in a range from 200 MHz to 1.2 GHz.

24. The method of claim 23 wherein the frequency range is from 300 MHz to 1 GHz.

25. The method of claim 24, wherein the frequency is about 500 MHz.

26. The method of any one of claims 1-25, wherein the T_1 experiment comprises magic angle spinning.

27. The method of any one of claims 1-26, wherein all of the T_1 experiments are conducted within a time period of up to 48 hours.

28. The method of claim 27, wherein all of the T_1 experiments are conducted within a time period of up to 24 hours.

29. The method of claim 28, wherein all of the T_1 experiments are conducted within a time period of up to 12 hours.

30. The method of claim 29, wherein all of the T_1 experiments are conducted within a time period of up to 6 hours.

31. The method of any one of claims 1-30, wherein macromolecule is a biologic molecule.

32. The method of claim 31, wherein the biologic molecule is a protein.

33. The method of claim 32, wherein the protein is an antibody or a bispecific antibody construct.

34. The method of any one of claims 1-33, wherein the solid state formulation is a lyophilized formulation.

35. The method of claim 34, wherein the fitting of step (f) is monoexponential.

36. The method of claim 34 or 35, wherein the highest temperature is in a range from 50 °C to 150 °C.

37. The method of claim 36, wherein the highest temperature is in a range from 100 °C to 150 °C.

38. The method of any one of claims 34-37, wherein the highest and lowest temperatures have a difference of at least 50 °C.

39. The method of any one of claims 34-38, wherein the T_1 experiment comprises a variable delay period in a range from 0.01 seconds to 60 seconds.

40. The method of any one of claims 1-10 or 12-33, wherein the solid state formulation is a frozen formulation.

41. The method of claim 40, wherein the fitting of step (f) is biexponential.

42. The method of claim 40 or 41, wherein the highest temperature is from -15 °C to 0 °C.

43. The method of claim 42, wherein the highest temperature is from -15 °C to -10 °C.

44. The method of any one of claims 40-43, wherein the highest and lowest temperatures have a difference of at least 40 °C.

45. The method of any one of claims 40-44, wherein each T_1 experiment comprises nine variable delay times at each temperature to generate nine FID plots at each temperature.

46. The method of any one of claims 40-45, wherein the T_1 experiment comprises a variable delay period in a range from 0.1 seconds to 240 seconds.

47. The method of claim 1, wherein the solid state formulation is a lyophilized formulation and:

the saturation recovery curve is generated by plotting the signal intensity of each of the at least three FID plots versus delay time;

the T_1 experiment comprises baseline suppression and magic angle spinning;

steps (d)-(f) are repeated at 15 or more temperatures;

each temperature is in a range from -50 °C to 150 °C;

the highest and lowest temperatures have a difference from 75 °C to 100 °C;

in each equilibrating step the solid state formulation is held at the temperature for a duration in the range of one minute to ten minutes before conducting the T_1 experiment;

the method excludes retuning and recalibrating the ssNMR probe after equilibrating at the first temperature;

the variable delay period is in a range from 0.1 seconds to 60 seconds; and

each T_1 experiment comprises six variable delay times at each temperature to generate six FID plots at each temperature.

48. The method of claim 1, wherein the solid state formulation is a frozen formulation and:

the saturation recovery curve is generated by plotting the signal intensity of each of the at least three FID plots versus delay time;

the T_1 experiment comprises baseline suppression and magic angle spinning;

steps (d)-(f) are repeated at 25 or more temperatures;

each temperature is in a range from -50 °C to 0 °C;

the highest and lowest temperatures have a difference from 25 °C to 40 °C;

in each equilibrating step the solid state formulation is held at the temperature for a duration in the range of one minute to ten minutes before conducting the T_1 experiment;

the method excludes retuning and recalibrating the ssNMR probe after equilibrating at the first temperature;

the variable delay period is in a range from 0.01 seconds to 240 seconds; and

each T_1 experiment comprises nine variable delay times at each temperature to generate nine FID plots at each temperature.

49. The method of claim 47 or 48 further comprising:

(g) converting each T_1 value to ^1H spin-lattice relaxation rate ("R_1"), and

(h) plotting R_1 versus temperature to generate a relaxation rate curve for the solid state formulation.

50. The method of claim 49, further comprising analyzing the relaxation rate curve to determine the molecular mobility of the macromolecule in the solid state formulation, the degree of aggregation in the solid state formulation, and/or the stability of the solid state formulation.

51. The method of any one of claims 47-50, wherein the macromolecule is a biologic molecule.

52. The method of claim 51, wherein the biologic molecule is a protein.

53. A method of selecting a macromolecule-containing solid state formulation among a group of test macromolecule-containing solid state formulations, the method comprising:

(I) generating a relaxation rate curve for each macromolecule-containing solid state formulation in the group of test macromolecule-containing solid state formulations; wherein the relaxation rate curve for each macromolecule-containing solid state formulation is generated by:

(a) equilibrating the solid state formulation at a first temperature;

(b) conducting a ^{1}H spin-lattice relaxation time ("T_1") experiment on the solid state formulation at the first temperature using direct detection ^{1}H ssNMR to generate at least three free induction decay ("FID") plots at the first temperature, wherein the T_1 experiment comprises baseline suppression and a saturation recovery sequence having at least three variable delay times from which each FID plot is generated;

(c) equilibrating the solid state formulation at a second temperature, and repeating step (b) at the second temperature to generate at least three FID plots at the second temperature;

(d) equilibrating the solid state formulation at a third temperature and repeating step (b) at the third temperature to generate at least three FID plots at the third temperature;

(e) generating a saturation recovery curve at each temperature;

(f) fitting each saturation recovery curve to a nonlinear regression equation to generate a T_1 value at each temperature;

(g) converting each T_1 value to ^{1}H spin-lattice relaxation rate ("R_1"), and

(h) plotting R_1 versus temperature to generate a relaxation rate curve for the solid state formulation;

(II) comparing the maximum R_1 peak value of each curve, the temperature of the maximum R_1 peak of each curve, the width of the maximum R_1 peak of each curve, or a combination thereof; and

(III) selecting the solid state formulation which has the lowest maximum R_1 peak value, the highest temperature of the maximum R_1 peak, or the narrowest R_1 peak width.

54. A method of selecting a formulation excipient for use in a macromolecule-containing solid state formulation, the method comprising:

(I) generating a relaxation rate curve for each macromolecule-containing solid state formulation in a group of test macromolecule-containing solid state formulations, each formulation having a different composition of excipients, a different amount of one or more excipients, or both; wherein the relaxation rate curve for each macromolecule-containing solid state formulation is generated by:

(a) equilibrating the solid state formulation at a first temperature;

(b) conducting a ^{1}H spin-lattice relaxation time ("T_1") experiment on the solid state formulation at the first temperature using direct detection ^{1}H ssNMR to generate at least three free induction decay ("FID") plots at the first temperature, wherein the T_1 experiment comprises baseline suppression and a saturation recovery sequence having at least three variable delay times from which each FID plot is generated;

(c) equilibrating the solid state formulation at a second temperature, and repeating step (b) at the second temperature to generate at least three FID plots at the second temperature;

(d) equilibrating the solid state formulation at a third temperature, and repeating step (b) at the third temperature to generate at least three FID plots at the third temperature;

(e) generating a saturation recovery curve at each temperature;

(f) fitting each saturation recovery curve to a nonlinear regression equation to generate a T_1 value at each temperature;

(g) converting each T_1 value to ^{1}H spin-lattice relaxation rate ("R_1"), and

(h) plotting R_1 versus temperature to generate a relaxation rate curve for the solid state formulation;

(II) comparing the maximum R_1 peak value of each curve, the temperature of the maximum R_1 peak of each curve, the width of the maximum R_1 peak of each curve, or a combination thereof; and

(III) selecting an excipient that is present in the solid state formulation that has the lowest maximum R_1 peak value, the highest temperature of the maximum R_1 peak, or the narrowest R_1 peak width.