Improved methods of manufacturing dexrazoxane that allow greater processing latitude are also provided.

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DEXRAZOXANE FORMULATIONS AND METHODS

FIELD OF THE INVENTION

The invention relates to pharmaceutical preparations and drug formulations of dexrazoxane.

BACKGROUND TO THE INVENTION

Dexrazoxane (ICRF-187) is a cardioprotective agent for use in conjunction with doxorubicin hydrochloride, an antineoplastic agent that is active against many forms of cancer. Dexrazoxane is an intracellular chelating agent that reduces or prevents myocardial toxicity associated with administration of doxorubicin HCl. Chemically, dexrazoxane is (S)-(+) bis-4,4’-(1-methyl-1,2-ethanediyl) 2,6-piperazmedione.

Commercially, dexrazoxane is currently formulated as a lyophilized product of a dexrazoxane hydrochloride salt (ZINECARD® or CARDIOXANE®). The product is available in 250 mg or 500 mg single use vials and is reconstituted in the clinic for intravenous injection. Reconstitution with 0.167 M sodium lactate injection, USP (co-marketed as diluent) results in an injectable solution of pH 3.5 - 5.5. Reconstitution using commonly available in-clinic infusion solutions of 0.9% sodium chloride solution or 5% dextrose in water solution results in a reconstituted solution of pH 1.4 - 1.8.

The current suggested human dose is about 2 g or four vials of 500 mg dose dexrazoxane drag product. Administration of a 2 g dose requires reconstitution and pooling of four 500 mg vials. In the clinic, this can be a time-consuming and laborious process. For example, dissolution of a 500 mg vial of lyophilized dexrazoxane HCl in sodium lactate requires approximately twenty minutes of shaking.

The commercialized lyophilized product is prepared by dissolving 20 mg/ml in 0.1 N HCl. The sterile filtered solution is filled into vials that are then placed in a pre-cooled lyophilizer set at 5°C. The time span from compounding to beginning freezing in a lyophilizer must not exceed 7 hours to prevent the decomposition of dexrazoxane in the formulation solution. This time limit puts a severe limitation on batch size.
It would be desirable to raise the pH of dexrazoxane infusate solutions to prevent local and systemic reactions to the low pH of infusate solutions. It would be further desirable to raise the pH of dexrazoxane injections and infusions to any pH desired for human administration.

It would be desirable to provide dexrazoxane in single unit dose containers (e.g. a 2g dexrazoxane container for a 2g dose). It would also be desirable to provide improved methods of preparation that allow manufacturers greater flexibility in providing multiple products suitable for different dosing ranges.

It would also be desirable to provide formulations and methods that reduce the amount of time in a clinic required to prepare the injectable solution. In particular, it would be desirable to eliminate the need to reconstitute and pool multiple vials of the lyophilized product.

It would further be desirable to provide methods of manufacturing in which batch size and tolerable process time may be increased.

**SUMMARY OF THE INVENTION**

The present invention meets these needs by providing improved formulations of dexrazoxane for making injectable solutions in a clinical setting. In some embodiments, high pH formulations are provided to reduce or eliminate pain upon injection. According to various embodiments, dexrazoxane may be formulated as a lyophilized or non-lyophilized acid salt or as a lyophilized or non-lyophilized free base. In some embodiments, dexrazoxane is provided as a sterile dry milled powder of a free base or an isolated acid salt crystal. Formulations that require significantly reduced clinical preparation time are provided. Single-container presentations of unit doses of dexrazoxane are also provided. The formulations of the present invention may be provided in a vial, infusion bottle or infusion bag. In some embodiments, the container may be directly attachable to a diluent-containing container for easy preparation. In other embodiments, diluent may be added directly to the container. Improved methods of manufacturing dexrazoxane that allow greater processing latitude are also provided.

One aspect of the invention relates to acid salt compositions of dexrazoxane prepared by dissolving at least 60 mg/ml of dexrazoxane in an acid solution. In some
embodiments, the compositions are prepared by dissolving at least 80 mg/ml or at least 100 mg/ml of dexrazoxane in the acid solution. According to various embodiments, the composition may be prepared by dissolving dexrazoxane in one of sulfuric, methanesulfonic, maleic, oxalic, phosphoric, and malonic acids. In certain embodiments, the composition may be prepared by dissolving dexrazoxane in one of sulfuric, methanesulfonic and maleic acids. In certain embodiments, the dexrazoxane salt of any of the above-described compositions is a bis, mono or sesqui-mono salt of dexrazoxane.

According to various embodiments, the acid salt composition may be an aqueous solution of the salt in acid, a lyophilized product, a reconstituted lyophilized product, or an isolated acid salt crystal. The aqueous solutions may be maintained at a temperature of at most about 50°C without precipitation, and in certain embodiments, the aqueous solution is stable for at least 48 hours, and in some embodiments, for at least 96 hours.

In certain embodiments, the acid salt compositions are lyophilized products, which may have less than 2% moisture. The lyophilized product compositions may also have greater than 500 mg dexrazoxane contained in a vial of no more than 50 ml.

The acid salt compositions include lyophilized products reconstituted in a solution for parenteral administration to a human. According to various embodiments, the concentration of dexrazoxane in the solution ranges from about 4 mg/ml to about 10 mg/ml. The reconstitution solution is one of an aqueous solution, a polar solution, a salt solution or an organic solution. For example, in certain embodiments, the reconstitution solution is a citrate-phosphate buffer having a pH ranging from 3 to 7 or having a pH ranging from 5.5 to 7.8. In certain embodiments, the reconstitution solution is a sugar solution, e.g., a sucrose or dextrose solution. The reconstitution solution may be selected from a sugar solution, sodium lactate, Ringers lactate, Plasma-Lyte A and sodium chloride. In certain embodiments, the reconstituted dexrazoxane is stable for at least 6 hours and in some embodiments, at least 8 hours.

According to various embodiments, the dexrazoxane salt of any of the above-described acid salt compositions may be amorphous or crystalline. In certain embodiments, the dexrazoxane salt is an isolated salt crystal. In certain embodiments,
any of the above-described acid salt compositions is substantially free of uncyclized analogs of dexrazoxane.

Also, in certain embodiments, the dexrazoxane salt of the acid salt composition is not a hydrochloric salt and/or not a sulfuric salt.

Another aspect of the invention relates to compositions containing pharmaceutically acceptable salts of dexrazoxane (ICRF-187) in solution. According to various embodiments, the composition has at least about 60 mg/ml dexrazoxane, at least about 80 mg/ml dexrazoxane or at least about 100 mg/ml dexrazoxane. According to various embodiments, the dexrazoxane salt of any of the above-described compositions is a bis, mono or sesqui-mono salt of dexrazoxane. Also according to various embodiments, the pharmaceutically acceptable salt of any of the above-described compositions is selected from a sulfate salt, a mesylate salt, a maleate salt, an oxalate salt, a malonate salt and a phosphate salt. In certain embodiments, the salt is selected from a sulfate salt, a mesylate salt and a maleate salt.

According to various embodiments, the composition of any of the above-described embodiments is an aqueous solution of the salt. The aqueous solution may be maintained at a temperature of at most about 50°C without precipitation. In certain embodiments, the above-described aqueous solutions are stable for at least 48 hours, and in some embodiments, for at least 96 hours. These compositions may be lyophilized products, which may have less than 2% moisture. In certain embodiments, the lyophilized product composition may have greater than 500 mg dexrazoxane contained in a vial of no more than 50 ml.

The above-described pharmaceutically acceptable salts of dexrazoxane (ICRF-187) in solution include lyophilized products reconstituted in a solution for parenteral administration to a human. According to various embodiments, the concentration of dexrazoxane in the solution ranges from about 4 mg/ml to about 10 mg/ml. The reconstitution solution is one of an aqueous solution, a polar solution, a salt solution or an organic solution. For example, in certain embodiments, the reconstitution solution is a citrate-phosphate buffer having a pH ranging from 3 to 7 or having a pH ranging from 5.5 to 7.8. In certain embodiments, the reconstitution solution is a sugar solution, e.g., a sucrose or dextrose solution. The reconstitution solution may be selected from a sugar solution, sodium lactate, Ringers lactate, Plasma-Lyte A and
sodium chloride. In certain embodiments, the reconstituted dexrazoxane is stable for at least 6 hours and in some embodiments, at least 8 hours.

In certain embodiments, the dexrazoxane salt of any of the above-described embodiments is amorphous or crystalline. In certain embodiments, the dexrazoxane salt is an isolated salt crystal. In certain embodiments, the composition of any of the above-described compositions is substantially free of uncyclized analogs of dexrazoxane.

Also, in certain embodiments, the composition of the above-described embodiments is not a hydrochloric salt and/or not a sulfuric salt.

Another aspect of the invention relates to aqueous solutions of acid salts of dexrazoxane having improved stability. In some embodiments, the aqueous solutions are stable for over 6 hours, for example having less than 3% total decomposition, at 25°C. Aqueous solutions of dexrazoxane are stable up to 96 hours at 5°C. Reconstituted lyophilized products stable up to 8 hours are also provided.

The lyophilized products of the invention may be presented in amounts greater than 500 mg, 750 mg, 1 g, or 2g in a single vial of no more than 50 ml. In certain embodiments, a unit dosage of at least 2 grams of a stable, lyophilized acid salt of dexrazoxane in a single vial of no more than 50 ml is provided.

Another aspect of the invention relates to lyophilized products of a mesylate (methanesulfonic), maleate, oxalate, phosphate and malonate salts of dexrazoxane. In some embodiments, these acid salts are prepared by dissolving at least 30 mg/ml of dexrazoxane in the acid. In some embodiments, the lyophilized products may be reconstituted in a solution. Lyophilized products reconstituted in a solution may be stable for at least 6 hours at about 25°C.

In certain embodiments, the acid salt is prepared by dissolving at least 30 mg/ml dexrazoxane in the acid, e.g., at least 30, 40, 60, 80 or 100 mg/ml. Any of the above-described lyophilized products may have less than 2% moisture. Also the lyophilized product compositions may have greater than 500 mg dexrazoxane contained in a vial of no more than 50 ml.

The lyophilized product may be reconstituted in a solution, e.g., for parenteral administration to a human. According to various embodiments, the concentration of
dexrazoxane in the solution ranges from about 4 mg/ml to about 10 mg/ml. In certain embodiments, the reconstitution solution is one of an aqueous solution, a polar solution, a salt solution or an organic solution. In certain embodiments, the reconstitution solution is a citrate-phosphate buffer having a pH ranging from 3 to 7 or having a pH ranging from 5.5 to 7.8. The reconstitution solution may be a sugar solution, e.g., a sucrose or dextrose solution. In certain embodiments, the reconstitution solution is selected from a sugar solution, sodium lactate, Ringers lactate, Plasma-Lyte A and sodium chloride.

In certain embodiments, the dexrazoxane compound of the lyophilized product reconstituted in a solution is stable for at least 6 hours, stability defined as having less or equal to 3% of the total decomposition product. In certain embodiments, the dexrazoxane compound is stable for at least 8 hours.

Another aspect of the invention relates to a unit dosage of at least 2 grams of a sterile pharmaceutical composition of a stable, lyophilized acid salt of dexrazoxane in a single vial of no more than 50 ml.

Another aspect of the invention relates methods of formulating acid salts of dexrazoxane as well as pharmaceutical compositions of said acid salts. In certain embodiments, the methods involve dissolving dexrazoxane in a solution comprising an acid of the salt. According to various embodiments from about 30 mg/ml - 100 mg/ml of dexrazoxane is dissolved in the acid solution. The acid may be selected from one of sulfuric, methanesulfonic, maleic, oxalic, phosphoric, and malonic acids. In certain embodiments, the acid is selected from one of sulfuric, methanesulfonic, maleic acids.

As indicated, methods of formulating pharmaceutical compositions are provided. In certain embodiments, the methods involve dissolving at least 60 mg/ml of dexrazoxane in a solution of an acid. In certain embodiments, at least 80 mg/ml of dexrazoxane is dissolved in the solution and in certain embodiments, at least 100 mg/ml of dexrazoxane is dissolved in the solution. The acid may be selected from one of sulfuric, methanesulfonic, maleic, oxalic, phosphoric, and malonic acids. In certain embodiments, the acid is selected from one of sulfuric, methanesulfonic and maleic acids. In certain embodiments, the acid is selected from one of methanesulfonic and sulfuric acids. The method may further involve filling sterile vials with a volume of
the acid salt solution to formulate a 2 gram dose of dexrazoxane in each vial. The vials may then be placed in a lyopholizer to create a lyophilized product having less than about 2% moisture content. The composition may be administered intravenously to a patient without substantial pain or local irritation at the site of administration.

Another aspect of the invention relates to methods of formulating pharmaceutical compositions that involve dissolving at least 30 mg/ml dexrazoxane (ICRF-187) in an acid at temperature less than or equal to about 25°C including refrigerated temperatures. According to various embodiments, the acid is selected from one of sulfuric, methanesulfonic, maleic, oxalic, phosphoric, and malonic acids.

In certain embodiments, the acid is selected from one of sulfuric, methanesulfonic and maleic acids. In certain embodiments, the acid is selected from one of methanesulfonic and sulfuric acids.

The methods of formulating pharmaceutical compositions may involve filling sterile vials with a volume of the solution. In certain embodiments, this volume is sufficient to formulate a 2 g dose of dexrazoxane in each vial. The methods may further involve placing the vials in a lyopholizer to create a lyophilized product having less than 2% moisture content.

In certain embodiments of the above-described methods of formulating pharmaceutical compositions, the composition is administered intravenously to a patient without substantial pain or local irritation at the site of administration.

Another aspect of the invention relates to a method of formulating a pharmaceutical composition that contains a salt of dexrazoxane for intravenous administration to a human. In certain embodiments, the method involves dissolving at least 60 mg/ml of dexrazoxane (ICRF-187) in solution comprising an acid of the salt; lyophilizing said solution comprising the acid of the salt to provide a lyophilizate; reconstituting the lyopliilizate in a single vial in a solution having a concentration of at least 60mg/mL; and diluting the reconstituted solution in an intravenous to a final concentration of 4mg/mL to 10mg/mL for administration to the human.

Another aspect of the invention relates to a cold-temperature manufacturing process. The process involves dissolving at least 30 mg/ml dexrazoxane in a solution comprising an acid at less than 25°C and in particular at 5°C. The cold-temperature
process allows for greater manufacturing latitude; in particular the process allows for a
pre-lyophilization time span of greater than 7 hours without significant
decomposition.

Another aspect of the invention relates to non-lyopholized sterile free base
dexrazoxane powders. According to various embodiments, the powder has a mean
particle size of less than 100 μm, 90 μm, 50 μm or 45 μm. In certain embodiments, at
least 80% of the particle size distribution is between 32 and 45 μm. The powder may
be dissolved in an infusion solution. According to various embodiments, the powder-
containing infusion solution may have a pH of at least 5.5, 6.0, 6.5 or 7.0 and may
have a pH greater than 6. The composition may contain at least one excipient to raise
pH. The powder of the above-described compositions may be amorphous or
crystalline. In certain embodiments, the powder has an X-ray diffraction pattern
having characteristic peaks at 2Θ (degree) of 8.4333, 12.6143, 13.5017, 14.8600,
15.2464, 16.1000, 16.4494, 16.8400, 17.8945, 18.4400, 18.7866, 19.3400, 22.4000,
22.6676, 24.2661, 25.1768, 25.4200, 27.1085, 27.9872, 29.3200, 29.6400, 31.3545,
31.7439, 32.2585, 32.8937, 34.8663, 35.3400, 37.8378, 38.5276, 39.0615 and 39.9800.

In certain embodiments, the above-described compositions of non-lyopholized
sterile free base powders are capable of being dissolved in an infusion solution in
under 5 minutes and/or may be capable of being fully dissolved in an infusion solution
prior to the decomposition of dexrazoxane. In certain embodiments, the infusion
solution is selected from a sugar solution, sodium lactate, Ringers lactate, Plasma-Lyte
A and sodium chloride. According to various embodiments, the pH of the powder-
containing solution is greater than 5 or 6. Also according to various embodiments, the
concentration of dexrazoxane in solution is between about 4 mg/ml to 10 mg/ml,
e.g., 4 mg/ml or 10 mg/ml.

In certain embodiments, the composition of the above-described non-
lyopholized sterile free base powders and powder-containing infusion solutions is
stable for at least 6 hours, and in certain embodiments, it is stable for at least 24 hours.

The above-described non-lyopholized free base powders may be packaged in
receptacles suitable for the addition of a buffer fluid to the powder. According to
various embodiments, the receptacles are configured for the addition of buffer fluid to
the powder or sterile transfer of the powder to a bag or bottle containing the buffer. Examples of receptacles include dockable vials, vials attached to infusion bags and IV infusion bottles.

Another aspect of the invention relates to packaged dexrazoxane products. The packaged product contains dry milled dexrazoxane (free base or acid salt crystals) in a receptacle suitable for the addition of a buffer. In some embodiments, the receptacle is configured for the addition of the buffer to the receptacle. In other embodiments, the receptacle is configured for the sterile transfer of the dexrazoxane to a bag or bottle containing the buffer. Examples of receptacles include a vial attached to an infusion bag, a dockable vial, an IV infusion bottle and an IV infusion bag.

Methods of formulating dexrazoxane pharmaceutical compositions are also provided. The methods involve dry milling dexrazoxane free base crystals or isolated acid salt crystals to form a dry milled powder. In some embodiments the dry milled powder is sieved to separate the particles into a desired size distribution. In some embodiments, dry milling the free base or acid salt involves using a ball mill or a tumula mixer.

Another aspect of the invention relates to formulating dexrazoxane (in clinic) for IV administration. The method involves dissolving dexrazoxane free base in an IV solution to a concentration of ≤ 10 mg/ml, e.g. by hand-shaking or machine agitation, in less than 5 minutes. In certain embodiments, the method further includes administering the formulation to a patient prior to the administration of doxorubicin. In certain embodiments, the free base is stable for at least 6 hours in the solution.

Another aspect of the invention relates to methods of counteracting cardiotoxicity during drug therapy for the treatment or prophylaxis of cancer. The methods involves administering a formulation of the invention to a patient. The formulation may be administered prior to the administration of an anthracycline. In embodiments wherein a free base formulation is administered to the patient, the administration is without the pain associated with low pH solutions. Other formulations described herein may be administered to a patient intravenously without substantial pain associated with injection of an acidic solution, particularly HCl.
In each of the aspects of the invention, dexrazoxane can be used in the manufacture of a medicament for treating or preventing cardiotoxicity, such as, cardiotoxicity associated with anthracycline administration, specifically doxorubicin administration.

Other embodiments provide the use of dexrazoxane prepared by any of the embodiments described herein, and another agent, such as an anthracycline, for simultaneous separate or sequential administration. In a more particular embodiment the other agent is doxorubicin. In another more particular embodiment the use is for treating cancer. In another embodiment the use is for treating cancer without cardiotoxicity.

Other embodiments provide a pharmaceutical preparation or system, comprising (a) dexrazoxane according to any of the aspects/embodiments described herein; and (b) an anthracycline, wherein said first and second agents are either in admixture or are separate compositions. In a more particular embodiment the second agent is doxorubicin. More specifically, the agents are for simultaneous separate or sequential administration. In another more particular embodiment the use is for treating cancer without cardiotoxicity.

These and other features of the invention will become more fully apparent when the following detailed description of the invention is read in conjunction with the accompanying drawings.

BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1 shows the percentage of decomposition products A, B and C present in the sulfate and mesylate salt forms of dexrazoxane and the reconstituted hydrochloric acid salt lyophilized dexrazoxane product after dilution to 10 mg/ml in Plasma-Lyte A, sodium lactate and Ringers lactate intravenous infusion fluids. Figure 1 provides a comparison of reconstitution stability of the dexrazoxane forms.

Figure 2 shows the percentage of decomposition products A, B and C present in the sulfate and mesylate salt forms of dexrazoxane and the reconstituted hydrochloric acid salt lyophilized dexrazoxane product after dilution to 4 mg/ml in Plasma-Lyte A,
Sodium lactate and Ringers lactate intravenous infusion fluids. Figure 2 provides a comparison of reconstitution stability of the dexrazoxane forms.

Figure 3 shows the percentage of decomposition products A, B and C present in the maleate, sulfate, mesylate salt forms of dexrazoxane and the reconstituted hydrochloric acid salt lyophilized dexrazoxane product at 25°C. Figure 3 provides a comparison of the pre-lyophilization processing stability of the dexrazoxane forms at 25°C.

Figure 4 shows the percentage of decomposition products A, B and C present in the maleate, sulfate, mesylate salt forms of dexrazoxane and the reconstituted hydrochloric acid salt lyophilized dexrazoxane product at 50°C. Figure 4 provides a comparison of the pre-lyophilization processing stability of the dexrazoxane forms at 50°C over 168 hours.

Figure 4A shows the percentage of decomposition products A, B and C present in the maleate, sulfate, mesylate salt forms of dexrazoxane and the reconstituted hydrochloric acid salt lyophilized dexrazoxane product at 50°C. Figure 4A provides a comparison of the pre-lyophilization processing stability of the dexrazoxane forms at 50°C over the 25 hours.

Figure 5 shows the percentage of decomposition products A, B and C present in dexrazoxane free base at 25°C at 4, 6, 8 and 24 hours after dilution with 5% Dextrose in Water, USP; 0.9% Sodium Chloride Solution, USP; Plasma-Lyte A Solution; Sodium Lactate Solution, USP; and Ringers’s Lactate Solution to 10 mg/ml.

Figure 6 shows the percentage of decomposition products A, B and C present in dexrazoxane free base at 25°C at 4, 6, 8 and 24 hours after dilution with 5% Dextrose in Water, USP; 0.9% Sodium Chloride Solution, USP; Plasma-Lyte A Solution; Sodium Lactate Solution, USP; and Ringers's Lactate Solution to 4 mg/ml.
Figure 7 shows the percentage of decomposition products A, B and C present in dexrazoxane free base at 50°C at 4, 6, 8 and 24 hours after dilution with 5% Dextrose in Water, USP; 0.9% Sodium Chloride Solution, USP; Plasma-Lyte A Solution; Sodium Lactate Solution, USP; and Ringer’s Lactate Solution to 4 mg/ml.

Figure 8 is a process flowsheet illustrating a process of manufacturing a sterile dry milled powder according to certain embodiments of the invention.

DESCRIPTION OF SPECIFIC EMBODIMENTS OF THE INVENTION

The materials and associated techniques of the present invention will now be described with reference to several embodiments. Important properties and characteristics of the described embodiments are illustrated in the structures in the text. While the invention will be described in conjunction with these embodiments, it should be understood that the invention is not intended to be limited to these embodiments. On the contrary, it is intended to cover alternatives, modifications, and equivalents as may be included within the spirit and scope of the invention as defined by the appended claims. In the following description, numerous specific details are set forth in order to provide a thorough understanding of the present invention. The present invention may be practiced without some or all of these specific details. In other instances, well known process operations have not been described in detail in order not to unnecessarily obscure the present invention.

As discussed above, dexrazoxane is a cardioprotective agent for use with doxorubicin. Commercially, dexrazoxane is currently formulated as a lyophilized acid salt of hydrochloride acid. The acid salt is unstable in aqueous solution and so is lyophilized for stability. To manufacture the commercial product, dexrazoxane is dissolved in 0.1 M HCl, sterile-processed and filled into vials. The vials are partially stoppered and loaded on the shelves of a lyophilization chamber. Compounding and vial-filling processes are carried out at ambient temperature. The solutions are frozen and the chamber evacuated until lyophilization is complete.
Typically, the solution occupies less than the full vial volume to optimize lyophilization time and to allow for expansion of the solution upon freezing. Current manufacturing techniques for Cardioxane®, a commercial dexrazoxane drug product involve placing 25 ml of solution is placed into 36 ml vials. The vials are then supplied to clinics in 500 mg doses. When needed, dexrazoxane is reconstituted in the clinic. A current recommended dose is 2g for an adult human. Thus, preparing a dose for injection previously required pooling four 500 mg doses. Increasing vial size is constrained by the conventional filling and the size of the lyophilization equipment, as well as drying time.

Dexrazoxane is subject to decomposition in aqueous solutions. Because of this, the time span from compounding to beginning of freezing in the lyophilizer must not exceed 7 hours without significant decomposition. Lyophilized dexrazoxane is stable for extended periods of time, on the order of 24 months without significant decomposition. Reconstituted dexrazoxane is stable for about 6 hours at room temperature.

The solubility of dexrazoxane in water at 25°C is about 10 mg/ml water. Dexrazoxane (ICRF-187) and its racemate (ICRF-159) are sparingly soluble in 0.1N HCl, slightly soluble in polar solvents such as ethanol and methanol and practically insoluble in non-polar, organic solvents [Repta, Baltezor and Bansal, J. Pharm Sci. 65 (1976) 238-242]. Generally speaking, the solubility of dexrazoxane increases as pH decreases. Formulating dexrazoxane as a salt of hydrochloric acid improves its solubility. Solubility of hydrochloric acid as high as 35 mg/ml in 0.1 N HCl at 25°C has been reported (U.S. Patent No. 4,963,551, hereby incorporated by reference), though therapeutic concentrations around 20-25 mg/ml are preferred (see U.S. Patent No. 5,760,039, hereby incorporated by reference). The concentration of dexrazoxane in the bulk solutions may also be quantified in terms of dexrazoxane molarity. A 35 mg/ml solution of dexrazoxane in 0.01 N HCl is a 0.13 M dexrazoxane (the molecular weight of dexrazoxane being 268.28 g/mol).

ACID SALTS OF DEXRAZOXANE

One aspect of the invention involves providing acid salts of dexrazoxane. As discussed above, the current pharmaceutical product is a lyophilized hydrochloric acid salt. The acid salts of the present invention provide improved solubility and stability.
over the hydrochloride acid salt. According to various embodiments, novel acid salts of sulfuric acid, methanesulfonic, maleic, oxalic and phosphoric acid (also referred to as sulfate, mesylate, maleate, oxalate and phosphate salts) are provided. In preferred embodiments, dexrazoxane salts of sulfuric acid, methanesulfonic and maleic acids are provided. The improved solubility and stability allow formulation of higher-concentration doses (in particular, a 2g dose may be provided in a single 36 ml or 50 ml vial or a vial of less volume as an 80 mg/ml or 100 mg/ml solution) and improved manufacturing processes.

The acid salts may be prepared by dissolving dexrazoxane in acids of varying concentrations. In some embodiments, a lyophilized product is then prepared. In other embodiments, the acid salt crystals are isolated.

Solubility

The acid salts of the present invention have improved solubilizing capacity over the current hydrochloric acid formulation. Higher concentration doses may be prepared. In order to present a 2g dose in a single lyophilized vial (of same size and methods of manufacture of the current 500 mg dose), a 0.30 M solution of dexrazoxane is required (80 mg/ml solubility, as much as 8 times the concentration currently used to make the 500 mg doses). In preferred embodiments, a 0.37 M solution of dexrazoxane (100 mg/ml solubility) is provided in order to formulate the 2g dose in a single lyophilized vial. A 100 mg/ml solution yields 2g in a 20 ml volume —allowing formulation of 2g dose in a single vial having a volume of 20 ml.

In some embodiments, acid salts of the present invention are prepared by dissolving at least about 80 mg/ml dexrazoxane in one of: sulfuric acid, maleic acid, methanesulfonic acid, oxalic acid and phosphoric acid.

In preferred embodiments, acid salts of the present invention are prepared by dissolving at least about 100 mg/ml dexrazoxane in one of sulfuric acid, maleic acid, methanesulfonic acid and oxalic acid.

The solubility of dexrazoxane in various inorganic acids was measured and is shown below in Table 1. Dexrazoxane was dissolved in 0.74 M and 0.56 M acid solutions, which are molar ratios of 2 and 1.5 acids to 1 dexrazoxane concentration (0.37 M). These concentrations theoretically yield in-situ bis and sesqui-mono salts of
dexrazoxane in solution. The solubility of the in-situ salts formed in solution is related to the acid strength (i.e., the dissociation constant pKa) and concentration of the acid and inversely related to pH. Solubility was measured at 25°C. Results are shown in Table 1.

Table I: Solubility of Dexrazoxane in Various Inorganic Acids

<table>
<thead>
<tr>
<th>Acids ID</th>
<th>Conc. (M)</th>
<th>Pka</th>
<th>Acid PH</th>
<th>Solubility (mg/mL)</th>
<th>Final PH</th>
<th>Relative Standard Deviation %</th>
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</thead>
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<tr>
<td>Sulfuric</td>
<td>0.74</td>
<td>-3.0</td>
<td>0.28</td>
<td>286.8</td>
<td>1.28</td>
<td>1.8</td>
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<td></td>
<td>0.56</td>
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<td>152.1</td>
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<td>1.5</td>
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<tr>
<td>Methanesulfonic</td>
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<td>177.5</td>
<td>1.66</td>
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<td>0.52</td>
<td>143.6</td>
<td>1.71</td>
<td>1.5</td>
</tr>
<tr>
<td>Oxalic</td>
<td>0.74</td>
<td>1.20</td>
<td>0.80</td>
<td>168.0</td>
<td>1.93</td>
<td>4.5</td>
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<td></td>
<td>0.56</td>
<td></td>
<td>0.87</td>
<td>128.2</td>
<td>1.92</td>
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<td>Maleic</td>
<td>0.50</td>
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<td>1.21</td>
<td>103.9</td>
<td>2.07</td>
<td>1.9</td>
</tr>
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<td></td>
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</tr>
<tr>
<td>Phosphoric</td>
<td>0.74</td>
<td>2.15</td>
<td>1.07</td>
<td>91.9</td>
<td>1.79</td>
<td>0.9</td>
</tr>
<tr>
<td></td>
<td>0.56</td>
<td></td>
<td>1.20</td>
<td>75.2</td>
<td>1.88</td>
<td>1.9</td>
</tr>
<tr>
<td>Malonic</td>
<td>0.74</td>
<td>2.88</td>
<td>1.53</td>
<td>61.7</td>
<td>2.20</td>
<td>1.0</td>
</tr>
<tr>
<td></td>
<td>0.56</td>
<td></td>
<td>1.66</td>
<td>49.6</td>
<td>2.25</td>
<td>1.5</td>
</tr>
<tr>
<td>HCl</td>
<td>0.74</td>
<td>-8.00</td>
<td>0.37</td>
<td>187.3</td>
<td>1.77</td>
<td>1.3</td>
</tr>
<tr>
<td></td>
<td>0.56</td>
<td></td>
<td>0.52</td>
<td>145.7</td>
<td>1.83</td>
<td>1.9</td>
</tr>
<tr>
<td></td>
<td>0.25</td>
<td></td>
<td>0.83</td>
<td>75.4</td>
<td>2.05</td>
<td>1.6</td>
</tr>
<tr>
<td></td>
<td>0.20</td>
<td></td>
<td>0.98</td>
<td>61.4</td>
<td>2.02</td>
<td>0.4</td>
</tr>
<tr>
<td></td>
<td>0.10</td>
<td></td>
<td>1.32</td>
<td>30.5</td>
<td>2.38</td>
<td>3.0</td>
</tr>
</tbody>
</table>

As shown in Table 1, four in-situ salt-forms - sulfate, methanesulfonate, oxalate and maleate - had a solubility of at least 100 mg/mL.

In some embodiments of the invention, the salt is a bis, mono or sesqui-mono salt of dexrazoxane. It has been found that accurately titrating the acid with equimolar amounts of acid to get mono or sesqui-mono or bis salt in-situ rather than arbitrary acid addition prevents the pH from dipping too low to unacceptable physiologic levels. It should be noted that HCl in Table 1, also shows solubility of at least 100 mg/ml at 0.56 M and 0.74 M, while maintaining a pH above 1.5. It would have been expected that the increasing the HCl acid concentration from the currently used concentration of 0.1 M to 0.56 or 0.74 M would also increase acidity by 5.6 or 7.4 fold, thereby greatly reducing pH of the final solution to a pH below 1. However, as shown in Table 1, the final pH for 0.74 M is 1.77 and for 0.56 is 1.83. This unexpected result shows that accurately titrating the acid with equimolar amounts of
acid to get mono or sesqui-mono or bis salt in-situ, rather than arbitrary acid addition, prevents the pH from dipping too low to unacceptable physiologic levels.

Table 2 shows the solubility of dexrazoxane in methanesulfonic, sulfuric and maleic acids at various concentrations at 5°C and 25°C.

Table 2: Solubility of dexrazoxane in methanesulfonic, sulfuric and maleic acids at 5°C and 25°C

<table>
<thead>
<tr>
<th>Temperature</th>
<th>Acid</th>
<th>Concentration (M)</th>
<th>Final pH</th>
<th>Solubility (mg/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Time (hr)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1</td>
</tr>
<tr>
<td>25°C</td>
<td>Methanesulfonic</td>
<td>0.25</td>
<td>1.86</td>
<td>68.0</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.40</td>
<td>1.68</td>
<td>106.1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.56</td>
<td>1.71</td>
<td>143.6</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.74</td>
<td>1.66</td>
<td>177.5</td>
</tr>
<tr>
<td></td>
<td>Sulfuric</td>
<td>0.25</td>
<td>1.66</td>
<td>99.8</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.40</td>
<td>1.55</td>
<td>146.7</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.56</td>
<td>1.36</td>
<td>152.1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.74</td>
<td>1.28</td>
<td>286.8</td>
</tr>
<tr>
<td></td>
<td>Maleic</td>
<td>0.25</td>
<td>2.22</td>
<td>61.0</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.40</td>
<td>2.11</td>
<td>74.7</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.50</td>
<td>2.07</td>
<td>103.9</td>
</tr>
<tr>
<td>5°C</td>
<td>Methanesulfonic</td>
<td>0.25</td>
<td>1.86</td>
<td>61.7</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.40</td>
<td>1.68</td>
<td>89.3</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.56</td>
<td>1.71</td>
<td>--</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.74</td>
<td>1.66</td>
<td>--</td>
</tr>
<tr>
<td></td>
<td>Sulfuric</td>
<td>0.25</td>
<td>1.66</td>
<td>84.3</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.40</td>
<td>1.55</td>
<td>123.5</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.56</td>
<td>1.36</td>
<td>--</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.74</td>
<td>1.28</td>
<td>--</td>
</tr>
<tr>
<td></td>
<td>Maleic</td>
<td>0.25</td>
<td>2.22</td>
<td>57.3</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.40</td>
<td>2.11</td>
<td>77.5</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.50</td>
<td>2.07</td>
<td>--</td>
</tr>
</tbody>
</table>
Dexrazoxane attained solubility of at least 100 mg/mL in 0.40M sulfuric acid and methanesulfonic acid solutions within one hour at 25°C. The final pHs of the solutions were pH 1.55 for sulfuric acid and pH 1.68 for methanesulfonic acid. These pH values are comparable to the pH of the present hydrochloric acid based dexrazoxane formulation (target pH = 1.5) but the solubilities of the sulfate and methanesulfonic salts allows the formulation of these salt-forms at a concentration four-fold that of the present formulation.

Thus, in some embodiments, acid salts of the present invention are prepared by dissolving at least about 80 mg/ml dexrazoxane in one of: sulfuric acid having a concentration of about 0.25 M, maleic acid having a concentration of about 0.5 M, methanesulfonic acid having a concentration of about 0.4 M, oxalic acid having a concentration of about 0.5 M or phosphoric acid having a concentration of about 0.7 M.

In preferred embodiments, acid salts of the present invention are prepared by dissolving at least about 100 mg/ml dexrazoxane in one of: sulfuric acid having a concentration of about 0.40 M, maleic acid having a concentration of about 0.5 M, methanesulfonic acid having a concentration of about 0.4 M or oxalic acid having a concentration of about 0.5 M. (Preparing a 100 mg/ml solution requires a concentration of about 0.25 M for sulfuric acid, about 0.38 M for methanesulfonic acid and about 0.49 M for maleic acid. Similarly 80 mg/ml requires a concentration of about 0.20 M for sulfuric acid, about 0.30 M for methanesulfonic acid and about 0.43 M for maleic acid.) However, as discussed above, in preferred embodiments, concentrations that yield in-situ bis, mono or sesqui-mono salts of dexrazoxane in solution are used.

In preferred embodiments, the resulting pH of the bulk solution prepared is about pH 1.55 for sulfuric acid and about pH 1.68 for maleic acid.

In some embodiments, the acid salts of the present invention may also have concentrations of dexrazoxane of around 0.11 M - 0.15 M (corresponding to 30 mg/ml - 40 mg/ml solubility or a 750mg - 1g dose). Solubility studies were performed at 50°C for one hour on 30 mg/mL and 40 mg/mL dexrazoxane solutions in
each of the solubilizing acids ranging in concentration from 0.05 M to 0.20 M. Methanesulfonic and sulfuric acids require a concentration of 0.1 M or higher to dissolve 30mg/mL dexrazoxane drug substance. At the minimum concentration of 0.1 M, the pH of the formulation is 2.14 for methanesulfonic acid and 1.64 for the stronger sulfuric acid. Maleic acid required a concentration of 0.125 M to dissolve 30 mg/mL and the formulation concentration pH was 2.19.

For a formulation containing 40 mg/mL dexrazoxane, a 0.1 M sulfuric acid or higher is required for complete drug dissolution and a minimum concentration of 0.15 M and 0.175 M is required for maleic acid and methanesulfonic respectively to dissolve 40mg/mL. The pHs of sulfuric, methanesulfonic and maleic acids following complete dissolution at these concentrations are 1.91, 1.85 and 2.26 respectively.

Thus, in some embodiments the invention involves dissolving about 30 mg/ml dexrazoxane in one of: methanesulfonic acid having a concentration of at least 0.1 M or maleic acid having a concentration of at least 0.125 M. In other embodiments, the invention involves dissolving about 40 mg/ml dexrazoxane in one of: maleic acid having a concentration of a least 0.15 M and methanesulfonic acid having a concentration of at least 0.175 M. Additionally, some embodiments involve dissolving 30—40 mg/ml in sulfuric acid having a concentration of at least 0.1 M at 50°C.

Although the above discussion focuses on dexrazoxane concentrations appropriate for 750 mg, 1g and 2g doses, the present invention encompasses other dexrazoxane concentrations. Given the details present herein for the solubility of dexrazoxane in sulfuric, methanesulfonic, maleic, oxalic and phosphoric acids of various concentrations, one of skill in the art would understand how to prepare an acid salt of a desired dexrazoxane concentration.

**Stability**

In some embodiments, the in situ acid salts of the present invention are lyophilized for reconstitution in a clinic. Stability of the drug is important at all stages: 1) during manufacturing/processing, 2) storage as a lyophilized product and 3) and in an intravenous infusion fluid. As with conventional dexrazoxane HCl
formulation, the lyophilized acid salts of the present invention are stable for relatively long periods of time in storage.

Figures 1 and 2 show the reconstitution stability of the sulfate and mesylate salt forms in three intravenous infusion fluids (Plasma-Lyte A, Sodium lactate and Ringers lactate), as compared with the current hydrochloric acid salt lyophilized product (reconstituted Cardioxane®). Samples were stored for 8 hours at 25°C after dilution. As explained below in Example 3, samples were analyzed with HPLC, and the percentage of total peak area made up of decomposition product peaks A, B and C was determined. This \( \% (A + B + C) \) is shown on the y-axis; the most stable salts have the lowest percentage. A salt is considered stable if the percentage of decomposition products is less than 3%. Figure 1 shows stability after dilution to 10 mg/ml and Figure 2 after dilution to 4 mg/ml.

As shown in Figures 1 & 2, all three salt forms are stable at both concentrations up to 8 hours at 25°C in all three infusion fluids. Stability of the sulfate and mesylate forms in the infusion fluids are in most cases superior to that of the lyophilized hydrochloric acid salt. Maleic acid would be expected to have superior stability as well.

It should also be noted that the highest pH obtained was for mesylate salt dissolved in sodium lactate (4.68) - comparable to the current lyophilized product dissolved in sodium lactate (4.66).

Additional studies showed that at 25°C, none of the salt forms were stable for 24 hours in any infusion fluid at either concentration. At 5°C, all four salt forms were stable up to 24 hours at both concentrations in all three infusion fluids.

Figures 3 and 4 compare the processing stability of maleate, sulfate and mesylate salt forms with the lyophilized hydrochloride salt form. Figure 3 shows the stability at 25°C. The maleate, sulfate and mesylate salt forms were stable up to 6 hours at 25°C and show better stability than the hydrochloride salt form, in some embodiments, a salt form is considered to have processing stability if the percentage of the total peak area is less than 1% peak A, less than 1% peak B and less than 0.5% peak C. This standard is met by the maleate, sulfate and mesylate forms at 6 hours, whereas decomposition the lyophilized hydrochloric acid salt form exceeded this standard at 2 hours. The processing stability specification limit is: \( (A+B+C) \leq 2.5\% \) or
A \leq 1.0\%, B \leq 1.0\% and C \leq 0.5\%. Chemical structures of A, B and C are given below in Example 3. Figures 4 and 4A shows processing stability at 5^\circ\text{C} (Figure 4 shows the data plotted over 168 hours and Figure 4A shows the data plotted over 25 hours.) At 5^\circ\text{C}, all four salt forms were stable up to 48 hours while the mesylate salt was stable up to 96 hours. At both 5^\circ\text{C} and 25^\circ\text{C}, maleate, sulfate and mesylate salts are more stable than the hydrochloric acid salt, with mesylate being most stable.

Manufacturing

As discussed above, the conventional processes for compounding dexrazoxane HCl and vial filling processes are performed in ambient temperatures. At this temperature, the compounding and vial filling must be completed within 6 - 7 hours to minimize the decomposition of the active drug. The acid salts are more stable at lower temperatures, however, refrigeration is difficult with hydrochloric acid as dexrazoxane is less soluble and will precipitate at these temperatures.

According to various embodiments, the present invention provides methods of preparing lyophilized acid salt products that allow pre-lyophilization process times over 24 hours. In some embodiments, the invention provides methods of preparing lyophilized acid salt products wherein the pre-lyophilization processes (i.e. compounding and vial-filling) occur at refrigeration temperatures, e.g. at about 5^\circ\text{C}.

As discussed above, the acid salts of the present invention are more soluble than the hydrochloric acid salt. Referring back to Table 1, the solubility of dexrazoxane at 25^\circ\text{C} in 0.1 M hydrochloric acid solution is 30.5 mg/mL. Identical concentrations of the sulfuric and methanesulfonic acids are readily solubilized at the refrigeration temperature of 5^\circ\text{C}. At 5^\circ\text{C}, a 30 mg/mL HCl solution precipitates from solution. The practical application of the higher solubilizing capacity of the acids of the present invention is that dexrazoxane formulation can be compounded in jacketed tank with the option to cool the tank to a low temperature, e.g. 5^\circ\text{C}, by circulating cold water through the jacketed tank without dexrazoxane precipitating from solution. The longer allowable processing time allows more vials to be filled and increases batch size.
In addition, the greater stability of the acid salts of the present invention increase the maximum processing time, as compared to the hydrochloride acid salt (from 6-7 to over 24 hours).

Isolated Acid Salt Crystals

As discussed above, the aqueous salt solutions of dexrazoxane may be lyophilized to provide as a stable, lyophilized product. In alternate embodiments, the dexrazoxane is formulated as an isolated crystal of the sulfate, mesylate, maleate, oxalate or phosphate salts.

The isolated crystals may be formed by any known method including cooling, anti-solvent, solvent extraction and direct precipitation methods. Crystalline forms characterized by the X-ray powder diffraction (XRPD) data shown in Table 2A were formed according to these methods.

Table 2A: Acid Salt Crystals

<table>
<thead>
<tr>
<th>Compound</th>
<th>Crystallization Solvent</th>
<th>Crystallization Method</th>
<th>2θ of three highest peaks (degrees)</th>
</tr>
</thead>
<tbody>
<tr>
<td>ICRF-187</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>free base</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>dioxane, water</td>
<td>Solvents Evaporation</td>
<td></td>
<td>7.4590, 19.1168, 25.0873</td>
</tr>
<tr>
<td>ICRF-187</td>
<td>dioxane, water</td>
<td>Solvents Evaporation</td>
<td>18.9051, 15.2111, 19.5444</td>
</tr>
<tr>
<td>sulfate</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ICRF-187</td>
<td>dioxane, water</td>
<td>Solvents Evaporation</td>
<td>18.9655, 15.2677, 25.2508</td>
</tr>
<tr>
<td>mesylate</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ICRF-187</td>
<td>dioxane, methanol</td>
<td>Cooling</td>
<td>19.6054, 25.5769, 15.2823</td>
</tr>
<tr>
<td>sulfate</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ICRF-187</td>
<td>dioxane, methanol</td>
<td>Cooling</td>
<td>29.8588, 21.0471, 19.2702</td>
</tr>
<tr>
<td>mesylate</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ICRF-187</td>
<td>Dioxane</td>
<td>Direct precipitation</td>
<td>18.8571, 15.7169, 25.1021</td>
</tr>
<tr>
<td>sulfate</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ICRF-187</td>
<td>Dioxane</td>
<td>Direct precipitation</td>
<td>7.5292, 22.4388, 19.1664</td>
</tr>
<tr>
<td>mesylate</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>sulfate</td>
<td>acetonitrile, wash with</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Ethyl acetate</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ICRF-187</td>
<td>N,N- dimethylformamide,</td>
<td>Anti-solvent</td>
<td>19.5671, 25.5341, 9.7707</td>
</tr>
<tr>
<td>mesylate</td>
<td>acetonitrile, wash with</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Ethyl acetate</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
In some embodiments, the invention provides crystalline forms of dexrazoxane salts as characterized by having peaks at or about the 2Θ angles indicated in Table 2A. For example, the first dexrazoxane sulfate salt in Table 2A may be characterized as having peaks at or about 7.46, 19.12, 25.09. In some embodiments, the crystalline forms are characterized by having peaks at the indicated 2Θ +/- 0.2 degrees.

The isolated salt crystals may be provided to a clinic as a sterile powder. Preparation of a sterile powder as discussed below with regard to a free base formulation may be applied to the isolated acid salt crystals. Alternatively, the isolated salt crystals may be supplied to a manufacturer for dissolution in water and subsequent lyophilization. This allows the manufacturer to prepare a lyophilized acid salt product without having to handle acids.
Pharmaceutical Compositions


FREE BASE DEXRAZOXANE

Another aspect of the invention involves providing dexrazoxane in a free base form. Free base formulations allow preparation of an injectable solution of high pH. Pain to the patient is thus eliminated or minimized. As discussed above, pHs of the acid salt forms are around 1 - 2. Higher pHs may be obtained by addition of the acid salts to a buffer - as shown in Figures 3 and 4, dilution with an infusion fluid raises the pH to between 4 and 5; and a pH of 3.1 - 5.5 has been reported for lyophilized dexraoxane HCl reconstituted with sodium lactate (Zinecard®).

However, it would be desirable to provide a dexrazoxane product that may be consistently used to make infusion solutions with a pH of at least 5, more preferably
greater than 5.5, even more preferably greater than 6. It would further be desirable to provide a dexrazoxane product at any desired pH.

According to various embodiments of the invention, dexrazoxane free base may be provided as a dry milled product or as a lyophilized product.

Sterile Dry Milled Free Base

In some embodiments, dexrazoxane free base is dry milled powder. The dry milled powders of the present invention have high rates of dissolution, making them practical for use in a clinical setting. According to various embodiments, dry milled powders of various mean particles sizes are provided. For example, in certain embodiments, mean particle sizes may be less than or equal to about 20, 32, 45, 63, 90, 125 or 180 µm. Generally, mean particles sizes around 50 µm are preferred.

The free base powder of the present invention is prepared by milling dexrazoxane. Dry milling may be accomplished by using standard milling procedures, for example, using a ball mill or a turbula mixer. In preferred embodiments, the milled powder is then sieved to separate the powder into the desired size distributions.

Particle Sizes

In preferred embodiments, the particle size of the powder is small enough to provide relatively high rates of dissolution of the powder in an infusion solution. Tables 3-6 show the particles size distribution (PSD) of dexrazoxane prior to milling, after milling for one hour at 20 vibrations per second in a ball mill and after milling for 2 hours at 67 RPM in a turbula mixer. Both milling processes were conducted at ambient temperature:

<table>
<thead>
<tr>
<th>Dexrazoxane as received (prior to milling)</th>
<th>Particle Size (µm)</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Greater than 250</td>
<td></td>
<td>43.9%</td>
</tr>
<tr>
<td>180-250</td>
<td></td>
<td>25.3%</td>
</tr>
<tr>
<td>125-180</td>
<td></td>
<td>17.1%</td>
</tr>
<tr>
<td>90-125</td>
<td></td>
<td>9.0%</td>
</tr>
<tr>
<td>63-90</td>
<td></td>
<td>4.1%</td>
</tr>
<tr>
<td>45-63</td>
<td></td>
<td>0.6%</td>
</tr>
<tr>
<td>Less than 45</td>
<td></td>
<td>0.0%</td>
</tr>
<tr>
<td>After ball milling</td>
<td>125</td>
<td>45.9%</td>
</tr>
</tbody>
</table>
Approximately 86.3% of the pre-milled powder exhibited particle diameters greater than 125 µm, and 24.7% of the powder is equal to or less than 125 µm. Following the one-hour ball-milling process, the profile of the PSD of the powder shifts towards smaller particle size ranges and the particle size ranged between about 20 µm -> 125 µm. Only 45.9% of the particles compared to 86.3% of the unmilled powder are greater than 125 µm, and no particle was found to be greater than 250 µm.

The turbula mixing process involved placing the milling ball on a receptacle loaded onto a turbula mixer and operating the turbula mixer for two hours. Compared to the PSD of the unmilled powder, size reduction of the powder particles was also achieved in the turbula mixer but to a lesser extent than in the ball mill. About 66.7% (a number between that found for the unmilled and ball-milled powders) of the particles is greater than 125 µm. The particle size diameters ranged between about 45 µm to about 180 µm. No particle size was greater than 250 µm.

It should be noted that the powder exhibited aggregation and clumping due to electrostatic charge that developed in the powder during the milling process. Therefore, there may have been particle sizes less than than 20 µm generated that are not revealed by the PSD data tabulated in Table 3 since this aggregation prevented these particles from being measured. However, it is believed that particles as small as 5 µm could be obtained by discharging the electrostatic charge using static dischargers or working in a controlled environment set at a moderate humidity of about 50% relative humidity. Moisture is known to discharge electrostatic charges.

Further, increasing milling time further reduces particle size and creates a larger proportion of the smaller particle ranges. Thus, powder with substantially all

<table>
<thead>
<tr>
<th></th>
<th>90-125</th>
<th>63-90</th>
<th>45-63</th>
<th>32-45</th>
</tr>
</thead>
<tbody>
<tr>
<td>After turbula mixing*</td>
<td>180</td>
<td>46.4%</td>
<td>125-180</td>
<td>20.3%</td>
</tr>
<tr>
<td></td>
<td>90-125</td>
<td>17.5%</td>
<td>63-90</td>
<td>10.6%</td>
</tr>
<tr>
<td></td>
<td>45-63</td>
<td>4.0%</td>
<td>32-45</td>
<td>0.0%</td>
</tr>
</tbody>
</table>

* Percentages for turbula mixing do not add up to 100% because about 1.2% of the powder was lost through a crack on the cover of the sieve assembly during sieving studies.
particles below 45 µm is created. Separation of the milled powder to obtain a narrower or wider particle size range is also performed.

Rate of Dissolution

The solubility of dexrazoxane in water and in various common infusion solutions is tabulated in Table 3A.

Table 3A: Solubility of dexrazoxane in five infusion solutions and water

<table>
<thead>
<tr>
<th>Diluents ID</th>
<th>Diluents pH</th>
<th>Solubility mg/mL</th>
<th>ICRF-187 pH</th>
<th>RSD%</th>
</tr>
</thead>
<tbody>
<tr>
<td>5% Dextrose, USP</td>
<td>4.52</td>
<td>11.0</td>
<td>5.37</td>
<td>1.4%</td>
</tr>
<tr>
<td>0.9% NaCl, USP</td>
<td>5.38</td>
<td>10.8</td>
<td>5.41</td>
<td>3.2%</td>
</tr>
<tr>
<td>Ringers lactate, USP</td>
<td>6.30</td>
<td>10.9</td>
<td>6.18</td>
<td>0.6%</td>
</tr>
<tr>
<td>Na lactate, USP</td>
<td>6.51</td>
<td>10.2</td>
<td>6.43</td>
<td>1.4%</td>
</tr>
<tr>
<td>Purified Water</td>
<td>6.68</td>
<td>10.4</td>
<td>6.25</td>
<td>-</td>
</tr>
<tr>
<td>Plasma-Lyte A, USP</td>
<td>7.07</td>
<td>9.70</td>
<td>6.37</td>
<td>4.3%</td>
</tr>
</tbody>
</table>

The solubility of dexrazoxane in these aqueous-based solutions ranges from 9.7—11.0 mg/mL. In a clinical setting, the rate of dissolution should be as high as possible to rapidly achieve any desired concentration (the maximum allowable concentration being the solubility in solution shown in Table 3). The faster the dissolution of dexrazoxane, the easier it is to constitute a solution of the sterile dry powder of the drug in a clinical or hospital setting.

Tables 4 and 5 show the rates of dissolution for five different size ranges of powders. API refers to active pharmaceutical ingredient, i.e. the unmilled dexrazoxane.
Regardless of the dissolution solvent, the dissolution rate of the powders decreased the smaller the particle size range of the powder. The complete dissolution time for these powders were identical for each of the powder particle size range evaluated in both the 0.9% sodium chloride and 5% dextrose solution, except for the 45 - 63 µm particle size range samples wherein dissolution in the sodium chloride solution was faster.

The quantity of powder employed was sufficient to provide about 8-mg/mL solutions when completely dissolved. However, the solubility of dexrazoxane in water and various commercially available infusion solutions, which is summarized in Table 3, show that its solubility in various aqueous solvent and solutions range from 9.7 to 11 mg/mL. Note that the dissolution endpoint for the solution in Tables 4 and 5 was 8 mg/ml corresponded to the entire amount of dexrazoxane used in the dissolution study. However, dissolution endpoints corresponding to the solubilities shown in Table 3 could also be achieved, and would be expected to have similar rates as those shown in Figures 3 and 4. Thus, constitution of a dry sterile powder of the

<table>
<thead>
<tr>
<th>Method</th>
<th>Size</th>
<th>Concentration (mg/mL)</th>
<th>Solvent</th>
<th>Hand shaking (min)</th>
<th>Eye observation</th>
</tr>
</thead>
<tbody>
<tr>
<td>API</td>
<td>&gt;125-180 um</td>
<td>8.0</td>
<td>0.9% NaCl</td>
<td>20</td>
<td>Complete</td>
</tr>
<tr>
<td>Ball mill</td>
<td>&gt;90-125um</td>
<td>7.9</td>
<td>0.9% NaCl</td>
<td>15</td>
<td>Complete</td>
</tr>
<tr>
<td>Turbula mill</td>
<td>&gt;63-90</td>
<td>8.3</td>
<td>0.9% NaCl</td>
<td>10</td>
<td>Complete</td>
</tr>
<tr>
<td>Turbula mill</td>
<td>&gt;45-63</td>
<td>8.1</td>
<td>0.9% NaCl</td>
<td>7</td>
<td>Complete</td>
</tr>
<tr>
<td>Ball mill</td>
<td>&gt;32-45</td>
<td>8.4</td>
<td>0.9% NaCl</td>
<td>3</td>
<td>Complete</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Method</th>
<th>Size</th>
<th>Concentration (mg/mL)</th>
<th>Solvent</th>
<th>Hand shaking (min)</th>
<th>Eye observation</th>
</tr>
</thead>
<tbody>
<tr>
<td>API</td>
<td>&gt;125-180 um</td>
<td>8.1</td>
<td>5% Dextrose</td>
<td>20</td>
<td>Complete</td>
</tr>
<tr>
<td>Ball mill</td>
<td>&gt;90-125um</td>
<td>8.1</td>
<td>5% Dextrose</td>
<td>15</td>
<td>Complete</td>
</tr>
<tr>
<td>Turbula mill</td>
<td>&gt;63-90</td>
<td>8.0</td>
<td>5% Dextrose</td>
<td>10</td>
<td>Complete</td>
</tr>
<tr>
<td>Turbula mill</td>
<td>&gt;45-63</td>
<td>8.2</td>
<td>5% Dextrose</td>
<td>10</td>
<td>Complete</td>
</tr>
<tr>
<td>Ball mill</td>
<td>&gt;32-45</td>
<td>8.0</td>
<td>5% Dextrose</td>
<td>3</td>
<td>Complete</td>
</tr>
</tbody>
</table>
present invention may be accomplished in as little as 3 minutes of hand-shaking. Particles sizes of ≤ 50 µm are preferred.

Reconstitution or dissolution times greater than a minute are not uncommon in hospital settings. The dissolution times shown in Tables 4 and 5 are better than or within the realm of the reconstitution times found with high dose commercial peptide and protein drug products. (Typical reconstitution time for lyophilized monoclonal antibodies and the high dose peptide drug, Cubicin®, are longer than 20 minutes).

**pH**

A major advantage of using a dexrazoxane free base drug product is that pain is minimized due to the high pH of the infusion solution. The dry milled dexrazoxane free base powders of the present invention yield solutions of pH ranging from about pH 5.7 - 6.9 when diluted to 4 mg/mL with commercially available infusion solutions and pH 6.4– 6.8 when diluted to 10 mg/mL with the same infusion solutions.

Table 3 shows initial pH of the infusion solutions as received from the suppliers, ranked from lowest to highest. The final pH of the solutions at the end of the dexrazoxane solubility experiments generally mimicked the same order except for water, which is the only pure solvent studied. For solutions or solvents with initial pH above 6, the final pH decreased in the presence of dexrazoxane. In contrast, initial solution pH below 6 increased at the end of the study in the presence of dexrazoxane. The final pH of the solutions at their determined solubilities were above pH 5 and ranged from pH 5.37 to 6.37. Since pH were determined at concentrations of approximately 10 mg/mL, which is equivalent to the maximum infusion concentration that will be administered in clinical settings, clinical infusion solutions containing 10 mg/mL dexrazoxane are expected to exhibit similar pH values.

The sterile fill dry powder product yields higher solution pH upon dilution with infusion solutions than the infusion solutions produced by dissolving dexrazoxane acid salt forms (see pHs of less than 5 in Figures 1 and 2 for the acid salt compared to pHs over 5.37 for the free base).

Higher pHs may be obtained by using different buffer or infusate solutions.
Stability

Figures 5-7 show the stability of dexrazoxane freebase in five commonly available intravenous infusion fluids at concentrations of 10 mg/ml at 25°C (Figure 5) and 4 mg/ml at 25°C (Figure 6) and 25°C (Figure 7). The 10 mg/mL infusion solutions precipitated from solution at 2 hours when stored at the refrigeration temperature of 5°C.

Stability is measured as the sum of the decomposition products A, B and C in infusion solutions, with a solution considered stable if the sum is less than or equal to 3%. The horizontal dotted line in Figures 5-7, which corresponds to 3% total decomposition products, visually delineates or demarcates the stability specification limit for the decomposition products in the infusion solutions as a function of time.

Figure 5 shows that at 10-mg/mL, the 5% Dextrose and the Ringers lactate infusion solutions are stable for 8 hours. Except for the Plasma-Lyte A solution, all the infusion solutions are stable for at least 6 hours. Plasma-Lyte A is not a commonly used infusion solution typically available in most hospitals. Thus all the readily available infusion solutions of the free base were stable for at least 6 hours at 25°C. At 8 hours following the preparation of the infusion solutions, only the 5% dextrose and the Ringers Lactate solutions met the stability specification of ≤ 3% total decomposition products.

Stability of dexrazoxane is a function of pH - the higher the pH of the infusion solution, the faster the rate of degradation. The higher the dexrazoxane concentration (10 mg/mL versus 4 mg/mL), the higher the final pH of the infusion solution, but the less stable the solution is. As noted above, 10 mg/mL infusion solutions should not be refrigerated because they form a precipitate with time due to the lower solubility at 5°C.

At a concentration of 4 mg/mL dexrazoxane free base in various infusion solutions, all the solutions refrigerated at 5°C were stable for 24 hours (Figure 7), but the same infusion solutions were stable for only 6 hours when stored at 25°C (Figure 6). At 25°C, the 5% dextrose, 0.9% Sodium Chloride and the Sodium Lactate infusion solutions exhibited stability for 8 hours. The pH of all the infusion solutions of the free base varied from pH 5.68 - 6.88.
Figure 8 is a process flowsheet showing a manufacturing process for the sterile dry milled powder according to one embodiment of the current invention. In step 801, the active drug product (API) dexrazoxane is added to a sterile crystallizing solvent or solution, for example WFI, in a jacketed tank. In preferred embodiments, dissolution is aided by heat. In step 803, the solution is filtered through sterilizing filters in a class 100 sterile room. In a specific embodiment 0.22µm filters are used. However, one of skill in the art will understand that any accepted sterilizing process and specifications may be used. Step 805 involves lowering the temperature of the drug solution in a controlled way to room or refrigerated temperature to crystallize a sterile powder of the drug. The powder is then collected by filtration, tray-dried, and filled into vials in step 807. The particles are then milled in step 809. Milling may be performed by any accepted milling process, for example, by ball mill or a turbula mixer as discussed above. In some embodiments, the milled particles are separated to achieve a desired size distribution. After milling and/or separating, the particles are filled into sterile vials in step 811 and the vials are stoppered in step 813. All of these processes are performed in a class 100 room to maintain sterility. As discussed below, the powder may also be filled into infusion bags or bottles.

The manufacturing process of the sterile dry milled powder is thus substantially easier than the current lyophilized process—not requiring freeze-drying. In addition, stability of the drug in aqueous solution (pre-lyophilization) is not a concern as discussed below. Thus, batch size may be increased as desired.

In addition to stability of dexrazoxane in an infusion solution, the shelf stability of the sterile dry fill product is important. In some embodiments, the shelf life of the commercial sterile dry fill product is longer because the above process retains the crystallinity of dexrazoxane, whereas the current lyophilization process yields an amorphous powder for reconstitution. The stability of dexrazoxane API is greater than 48 months when stored at room temperature in double polyethylene bags. The current amorphous product has an expiry dating of 36 months. The sterile API fill of this invention yields a drug product with a shelf life equivalent to that of the non-sterile API (or drug substance).
Although the above discussion relates to milled free base powder, the isolated acid salt crystals may also be provided as a dry milled sterile powder. One of skill in the art would understand how to apply the details given with respect to the free base powder to the isolated acid salt crystals.

Pharmaceutical Compositions

In addition to the dry milled powder, the pharmaceutical compositions of the present invention may contain powder excipients added to obtain any desirable pH. Examples of excipients are buffering agents (i.e., an acid and its conjugate base or a base and its conjugate acid) such as Acetate, Benzoate, Gluconate, Glycerate, Lactate, Aconitate, Adipate, Ascorbate, Carbonate, Glutamate, Malate, Maleate Succinate, Tartarate, Citrate, EDTA (edetate), Phosphates. Other buffering agents are Ammonia, Amino Acids, Diethanolamine and Tromethamine (TRIS, THAM). The dry powder filled into vials, bottles or bags may contain other excipients or ingredients that enhances the solubility / dissolution and / or stability of the API.

Lyophilized Free Base

Dexrazoxane free base may also be provided as a lyophilized product. The pH of a 10 mg/ml compounding solution is expected to be near pH 6.25. The pH of the reconstituted injectable solution is thus significantly greater than the current lyophilized hydrochloric acid salt product. However, pre-lyophilization processing stability is a concern, as with the lyophilized acid salt product. Aqueous solutions of dexrazoxane are more stable at low pH. Because of the higher pH, the maximum allowable time between compounding and lyophilization of a 10 mg/ml free base solution would be about 4 hours (as compared to about 6 for the lyophilized hydrochloric acid salt).

Recourse to a lower temperature to enhance stability is not a viable option because the solubility of dexrazoxane decreases with temperature and precipitation of dexrazoxane from solution will occur if using a 10 mg/ml solution.

It should be noted that the National Cancer Institute (NCI) has successfully lyophilized a 10-mg/mL free base solution as a 250 mg vial presentation. The
manufacturing batch size is unknown, and if it were a small batch size, it would be technically feasible to compound, sterile filter and fill the batch within two hours. The NCI product has never been commercialized.

5 **STERILE DRY POWDER FILL**

Another aspect of the invention involves packaging the dexrazoxane dry milled powder (free base or acid salt) formulations in receptacles that allow for easy preparation of the infusion solution in a clinical setting. According to various embodiments, the sterile dry milled powder is provided in a dockable vial, a vial pre-attached to an infusion bag, or in an infusion bag or bottle.

These presentations allow for simpler and quicker infusion solution preparations than currently required, in addition, potential for contamination, mixing errors and needle injuries is reduced, in preferred embodiments, the product does not need to be co-marketed with a diluent to raise the pH, simplifying and minimizing preparation time in a clinical setting.

In some embodiments, dexrazoxane (and excipients) are filled as sterile dry powder into a specialized docking or dock-able vials such as Abbott's ADD-Vantage® vials or Baxter's ULTIMATE PLUS® or SOLOMIX® diluent container with a pre-attached vial. Dockable vials containing the product are attached to the diluent infusion container and a given volume (e.g., about 10 mL) of infusion liquid is added to the vial. The vial is shaken well and the resulting suspension is transferred to the infusion solution container. The process is repeated with an additional equal volume of infusion solution to ensure complete transfer of vial contents to the infusion solution. The resulting mixture is vigorously shaken or agitated until clear.

When dexrazoxane is formulated in a vial pre-attached to an infusion bag, the combination packaging system is commercialized as a ready-to-mix product and a given volume of infusion solution is transferred into the vial, shaken vigorously and the resulting solution or suspension is transferred into the larger solution in the infusion bag. The procedure is repeated to ensure complete transfer of the vial contents into the infusion solution. The resulting mixture in the bag is shaken or agitated until completely clear.
When dexrazoxane is formulated in an infusion bag or bottle, infusion liquid is injected through a port directly to the bag and if formulated in a bottle, infusion solution is introduced into the bottle via an infusion set spike through the stopper. The powders are dissolved by of the introduced infusion solutions or liquid. The bottles or bags are shaken vigorously or agitated until the infusion solution is visually clear.

Because infusion solution is added as needed, there is effectively no upper limit on the size of dose that may be presented in a single container. High doses of dexrazoxane can be presented in one unit package (vial, bottle or bag). In preferred embodiments, each container has a single unit dose (e.g. of 2g). This obviates the need to pool four 500 mg vials to dose an adult as is presently done with the current drug product. However, one of skill in the art would understand that doses of any size desired size may be formulated.

EXAMPLES

The following examples provide details illustrating aspects of the present invention. These examples are provided to exemplify and more clearly illustrate these aspects of the invention and are in no way intended to be limiting.

Example 1: Solubility of Dexrazoxane in Various Inorganic Acids

Dexrazoxane (Lot No7166301), was supplied by Chiron Corporation, Europe and used as received. High performance liquid chromatography (HPLC)-grade water, EDTA disodium, and methanol were used to prepare all mobile phases. Infusion solutions such as dextrose 5% in Water USP, 0.9% Sodium Chloride Injection USP, Sodium Lactate Injection USP, Lactated Ringers Solution USP and Plasma-Lyte A were purchased from Baxter Corporation, Deerfield, IL. All other chemicals and solvents were reagent grade and were used as received.

Instrumentation

HPLC solubility analyses of the solubility solution samples were performed using a Waters™ Alliance model 2695 system consisting of a PDA UV detector model 2996, a pump model 2695, and an autosampler. The data was acquired and
analyzed on the Empower™ chromatography data system. The raw data are stored in server EMVGED. Solution pH was measured using a Fisher Scientific Accumet AB 15 pH meter equipped with a Beckman PN 511063 glass electrode.

**HPLC Method**

HPLC analyses of dexrazoxane solution samples were conducted. The isocratic reverse-phase HPLC method employed a Higgins, Haicart, Targa - C₁₈ P/N TS-1546-C185, 5 μm, 120 angstrom column (4.6 mm x 150 mm) and a Higgins, Haicart, Targa - C₁₈, P/N HK-GUARD-FM guard column, and a mobile phase of EDTA di-sodium (0.5 mM)/ methanol (89/11 v/v). The UV detection wavelength was set at 215 nm and the detector offset was 50 mV. The autosampler temperature was set at 20°C and the column was set at the thermostatic temperature of 30°C. The injection volume, flow rate and run time were 10 μL, 1.0 mL/min and 12 minutes respectively. The linearity of the method was demonstrated in the range of 50 -160 pg compound injected using peak area (r² = 0.9996).

**Solubility Studies**

Owing to the instability of dexrazoxane in aqueous media, particularly at pH >3, equilibrium solubility studies could not be conducted for the majority of the solubility studies. Solubility studies were limited to shaking or agitating an excess dexrazoxane drug powder suspended in 2 mL of solvent or solution of interest for one hour at a constant temperature (4°C or 25°C). In a few cases, equilibrium solubility studies were conducted for 24 hours below pH 2 in sulfuric, maleic and methanesulfonic acid solutions. Each solubility data point is the mean of three determinations.

Samples of the suspension were clarified by filtration through a 0.2 μm Nylon (Pall) filters. The first filtered milliliter of solution was discarded and subsequent filtered solution was collected for assay by reversed-phase HPLC. All assayed solutions were quenched with 0.1N hydrochloric acid to minimize the decomposition of the filtered solution in sample vials placed on an autosampler ready to be analyzed. The quenched solutions were further diluted with sodium hydroxide solutions (0.10 M and 0.05 M) and 0.5 mM EDTA di-sodium di-hydrate solution to pH 3.0 and to within
the linearity range of the assay method before being assayed by HPLC as described in more details in the next paragraph.

Depending upon the anticipated solubility range of the dexrazoxane compound in a given solvent, an appropriate µL aliquot between 20 µL and 250 µL of filtered solution is transferred into a 25 mL or 50 mL containing 1000 µL of 0.1 M HCl and mixed well. Just prior to HPLC analysis, an additional 1000 µL of 0.05 M sodium hydroxide is added to the flask, mixed and the volume in the flask is adjusted to the 25 mL or 50 mL mark with 0.5 mM EDTA di-sodium di-hydrate solution to dilute the sample to within the linearity range of the assay method.

Results of the solubility of dexrazoxane in various inorganic acids are shown in Table 1.

Example 2: Determination of Concentrations of Methanesulfonic, Maleic and Sulfuric Acids that dissolve 30 mg/mL and 40 mg/mL Dexrazoxane Drug Substance at 5°C

Dexrazoxane was added to various concentrations of methanesulfonic, maleic and sulfuric acids at 5°C. The samples were observed for dexrazoxane dissolution. Final pH was also measured. The results appear below in Tables E1-E3:

Table E1: Dissolution of dexrazoxane in methanesulfonic acid at 5°C

<table>
<thead>
<tr>
<th>Concentration of Dexrazoxane (mg/mL)</th>
<th>Concentration of Methanesulfonic Acid (M)</th>
<th>Visual Observation of Clarity</th>
<th>Final Solution pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>30 mg/mL</td>
<td>0.050</td>
<td>Incomplete Dissolution</td>
<td>2.44</td>
</tr>
<tr>
<td></td>
<td>0.100</td>
<td>Complete Dissolution</td>
<td>2.14</td>
</tr>
<tr>
<td></td>
<td>0.125</td>
<td>Complete Dissolution</td>
<td>1.83</td>
</tr>
<tr>
<td></td>
<td>0.150</td>
<td>Complete Dissolution</td>
<td>1.61</td>
</tr>
<tr>
<td></td>
<td>0.175</td>
<td>Complete Dissolution</td>
<td>1.35</td>
</tr>
<tr>
<td></td>
<td>0.200</td>
<td>Complete Dissolution</td>
<td>1.29</td>
</tr>
<tr>
<td>40 mg/mL</td>
<td>0.050</td>
<td>Incomplete Dissolution</td>
<td>2.47</td>
</tr>
<tr>
<td></td>
<td>0.100</td>
<td>Incomplete Dissolution</td>
<td>2.25</td>
</tr>
</tbody>
</table>
Table E3: Dissolution of dexrazoxane in maleic acid at 50°C

<table>
<thead>
<tr>
<th>Concentration of Dexrazoxane (mg/mL)</th>
<th>Concentration of Maleic Acid (M)</th>
<th>Visual Observation of Clarity</th>
<th>Final Solution pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.125</td>
<td>Complete Dissolution</td>
<td></td>
<td>2.15</td>
</tr>
<tr>
<td>0.150</td>
<td>Complete Dissolution</td>
<td></td>
<td>1.91</td>
</tr>
<tr>
<td>0.175</td>
<td>Complete Dissolution</td>
<td></td>
<td>1.64</td>
</tr>
<tr>
<td>0.200</td>
<td>Complete Dissolution</td>
<td></td>
<td>1.48</td>
</tr>
</tbody>
</table>

Table E2: Dissolution of dexrazoxane in sulfuric acid at 5°C

<table>
<thead>
<tr>
<th>Concentration of Dexrazoxane (mg/mL)</th>
<th>Concentration of Sulfuric Acid (M)</th>
<th>Visual Observation of Clarity</th>
<th>Final Solution pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>30 mg/mL</td>
<td>0.050</td>
<td>Incomplete Dissolution</td>
<td>2.35</td>
</tr>
<tr>
<td></td>
<td>0.100</td>
<td>Complete Dissolution</td>
<td>1.64</td>
</tr>
<tr>
<td></td>
<td>0.125</td>
<td>Complete Dissolution</td>
<td>1.45</td>
</tr>
<tr>
<td></td>
<td>0.150</td>
<td>Complete Dissolution</td>
<td>1.34</td>
</tr>
<tr>
<td></td>
<td>0.175</td>
<td>Complete Dissolution</td>
<td>1.10</td>
</tr>
<tr>
<td>40 mg/mL</td>
<td>0.050</td>
<td>Incomplete Dissolution</td>
<td>2.34</td>
</tr>
<tr>
<td></td>
<td>0.100</td>
<td>Complete Dissolution</td>
<td>1.85</td>
</tr>
<tr>
<td></td>
<td>0.125</td>
<td>Complete Dissolution</td>
<td>1.61</td>
</tr>
<tr>
<td></td>
<td>0.150</td>
<td>Complete Dissolution</td>
<td>1.45</td>
</tr>
<tr>
<td></td>
<td>0.175</td>
<td>Complete Dissolution</td>
<td>1.19</td>
</tr>
</tbody>
</table>
Example 3: Stability of Sulfate, Mesylate and Maleic Acid Salts

Materials and Chemicals

Dexrazoxane (ICRF-187) free base (Lot Number 7166301), was supplied by Chiron Corporation, Europe and was used as received. Maleic acid, sulfuric acid and methanesulfonic acid were purchased from Sigma-Aldrich. Cardioxane® (Batch #04D01-2) was supplied by Chiron Corporation, Europe as a lyophilized hydrochloride salt of dexrazoxane. High performance liquid chromatography (HPLC)-grade water, EDTA disodium, and methanol were used to prepare all mobile phases.

Infusion solutions such as dextrose 5% in Water USP, 0.9% Sodium Chloride Injection USP, Sodium Lactate Injection USP, Lactated Ringers Solution USP and Plasma-Lyte A were purchased from Baxter Corporation, IL. All other chemicals and solvents were reagent grade and were used as received.

Instrumentation

HPLC analyses of the stability samples were performed using a Waters™ Alliance model 2695 system consisting of a PDA UV detector model 2996, a pump model

<table>
<thead>
<tr>
<th>Concentration (mg/mL)</th>
<th>Dissolution State</th>
<th>Measured Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.150</td>
<td>Complete Dissolution</td>
<td>2.15</td>
</tr>
<tr>
<td>0.175</td>
<td>Complete Dissolution</td>
<td>2.03</td>
</tr>
<tr>
<td>0.200</td>
<td>Complete Dissolution</td>
<td>1.94</td>
</tr>
<tr>
<td>40 mg/mL</td>
<td>Incomplete Dissolution</td>
<td>2.61</td>
</tr>
<tr>
<td>0.100</td>
<td>Incomplete Dissolution</td>
<td>2.38</td>
</tr>
<tr>
<td>0.125</td>
<td>Incomplete Dissolution</td>
<td>2.32</td>
</tr>
<tr>
<td>0.150</td>
<td>Incomplete Dissolution</td>
<td>2.31</td>
</tr>
<tr>
<td>0.175</td>
<td>Complete Dissolution</td>
<td>2.26</td>
</tr>
<tr>
<td>0.200</td>
<td>Complete Dissolution</td>
<td>2.11</td>
</tr>
</tbody>
</table>
2695, and an auto sampler. The data on this system were collected, and processed using a personal computer equipped with the Empower™ software. Solution pH were measured using a Thermo Orion model 920 pH meter equipped with a Thermo Orion BN 8203 glassed electrode.

5 HPLC Method
Analyses of dexrazoxane stability samples were conducted using a strong cation exchange (SCX) HPLC method.

The chemical structures of A, B and C are given below:

![Chemical Structures](image)

Stability Studies

15 Stability studies were performed on the following samples:

1. Maleate, Sulfate and Mesylate salts of dexrazoxane
2. Hydrochloride salt of dexrazoxane (current lyophilized product)
3. Mesylate and Sulfate salts of dexrazoxane diluted with infusion fluids
4. Hydrochloride salt of dexrazoxane (current lyophilized product) diluted with infusion fluids

The stability studies were performed at 25°C and 5°C.

25 Stability solution preparations

1. Preparation of maleate, sulfate and mesylate (methanesulfonate) salts of dexrazoxane
Dexrazoxane (ICRF-187) free base was dissolved at a concentration of 30 mg/mL in 0.2M maleic acid, 0.2M sulfuric acid, or 0.2M methanesulfonic acid to form maleate, sulfate or mesylate salts in situ. Dexrazoxane dissolved in all three acids in about 5 minutes. Upon complete dissolution, the pH of each solution was measured and recorded. Lyophilized Cardioxane (hydrochloride salt) reconstituted with WFI to a concentration of 20 mg/mL was used as a control. One mL of each salt solution was stored at 25°C or 5°C. Stability samples were collected at 0, 2, 4, 6, 8, and 24 hours. Each sample was diluted with HPLC mobile phase to a concentration of about 1.0 mg/mL for HPLC analysis.

2. Preparation of mesylate and sulfate salts and lyophilized Cardioxane in three intravenous infusion fluids

Dexrazoxane (ICRF-187) freebase was dissolved at a concentration of 30 mg/mL in 0.15M methanesulfonic acid or 0.1M sulfuric acid to form mesylate or sulfate salts in situ. Dexrazoxane dissolved in both acids in about 2 minutes. The mesylate and sulfate salts were then diluted with Plasma-Lyte A, Sodium Lactate or Ringers lactate infusion fluids to concentrations of 10 mg/mL and 4 mg/mL. Lyophilized Cardioxane was reconstituted directly with Plasma-Lyte A, Na Lactate or Ringers lactate to concentrations of 10 mg/mL and 4 mg/mL. Upon complete dissolution or reconstitution, the pH of each solution was measured and recorded. The solutions were stored at 25°C or 5°C. Stability samples were collected at 0, 2, 4, 6, 8, and 24 hours. Each sample was diluted with HPLC mobile phase to a concentration of about 1.0 mg/mL for HPLC analysis.

Results

Processing Stability: Stability of maleate, sulfate, mesylate and hydrochloride (Lyophilized Cardioxane) salt forms: Stability of maleate, sulfate, mesylate and hydrochloride salt forms of dexrazoxane was evaluated at 50°C and 25°C and is listed in Tables 1 & 2. All three salt forms i.e. maleate, sulfate and mesylate were stable up to 6 hours at 25°C and were better than the hydrochloride salt form (Lyophilized Cardioxane). (Stability is defined as processing stability i.e. <1% peak A, <1% peak B
and <0.5% peak C.) At 50°C, all four salt forms were stable up to 48 hours while the mesylate salt was stable up to 96 hours. Results are shown in Figures 3 and 4.

Reconstitution Stability: Stability of maleate, sulfate, mesylate and hydrochloride salt forms of dexrazoxane was studied at 50°C and 25°C after dilution with three intravenous infusion fluids (Plasma-Lyte A, Na lactate and Ringers lactate) to concentrations of 4 & 10 mg/mL and compared to the stability of reconstituted lyophilized dexrazoxane in the same fluids. Results are shown in Figures 1 & 2, all four salt forms were stable at both concentrations up to 8 hours at 25°C in all three infusion fluids. Stability is defined as reconstitution stability i.e. < 3% total degradants: peak A + peak B + peak C. At 25°C, none of the salt forms were stable for 24 hours in any infusion fluid at either concentration. At 50°C, all four salt forms were stable up to 24 hours at both concentrations in all three infusion fluids. The highest pH obtained in this study at 4 & 10 mg/mL concentrations were with the current lyophilized product (hydrochloride salt) and the mesylate salt in Na lactate buffer (Figures 1 and 2).

**Example 4: Milling of Dexrazoxane Free Base**

ICRF-187 (Dexrazoxane, CAS RN 24584-09-6) was provided from Zinecard, Europe. The infusion solutions such as dextrose 5% in water USP, 0.9% sodium chloride USP, Plasma-Lyte A USP, sodium lactate USP and Ringers lactate USP were purchased from Baxter Corporation, IL. High performance liquid chromatography (HPLC)-grade water, acetonitrile, methanol, disodium EDTA, disodium phosphate were used to prepared all mobile phases. All other chemicals and solvents were reagent grade and were used as received.

**Instrumentation/Equipment:**

Dexrazoxane dry powder was milled using either a Ball Mill equipped with a 25mL stainless steel container and a stainless steel grinding ball (Mixer Mills, model MM301, Retsch GmbH & Co KG, Germany), or a Turbula Mixer (Model T2F, GlenMills, hie, NJ) loaded with a 50mL high-density polyethylene bottle containing a stainless steel grinding ball.
Milling studies were conducted in a Ball Mill or in a Turbula Mixer. For studies carried out in the ball mill, 12 grams of dexrazoxane powder was weighed into a 25 mL receptacle. A stainless steel grinding ball was then placed inside the receptacle. The receptacle was sealed and loaded onto the ball mill. The ball mill was horizontally shaken at a speed of 20 vibrations per second for one hour. For milling studies using the Turbula Mixer, 12 grams of dexrazoxane powder was added to a 50 mL high-density polyethylene bottle containing a stainless steel ball mill. The turbula mixer was then tumbled at 67 RPM for 2 hours. During shaking in the ball mill or tumbling in the turbula mixer, the stainless steel ball pulverizes the powder to smaller particle sizes. Both milling processes were conducted at ambient temperature.

The ground freebase from the ball mill was sieved with six sieves stacked from the largest sieve diameter to the smallest sieve diameter. The stacking order was 125, 90, 63, 45, 32, and 20 µm sieve meshes. For the turbula mixer, the stacking order was: 180, 125, 90, 63, 45, and 32 µm sieve meshes. The unmilled drug substance was also sieved to segregate the particles into different size distributions to serve as a control experiment. The stacking order for the control experiment using unmilled drug substance (also called active pharmaceutical ingredient) was: 250, 180, 125, 90, 63, and 45 µm. Sieving which comprises the automatic shaking and vibration of the stacked sieves was initiated by setting the pulse vibration/oscillation amplitude to 8 - 9. The shaking/vibration duration varied from 10 to 70 minutes depending upon the degree of pulverization desired for the particles in the mills. The sieved samples were collected in a high-density polyethylene bottle for further studies. The material balance calculation for the recovery of the powder from the various sieves after the sieving process was 98.7% to 100%.

Ground drug powder was sieved to separate the powders into different narrower size distributions using a Sonic Sifter Separator (Model L3P, Adantech Manufacturing, Inc., WT) equipped with stacked sieves (i.e. stacked in six levels). The mesh sizes of the sieves ranged from 20 to 250 µm. Sieve sizes were selected based upon the anticipated powder particles size distribution.

The resulting particle size distributions (PSDs) of the unmilled free base as received, after ball milling for an hour and after milling with the Turbula mixer are shown in Table 3.
**Example 5: Rate of Dissolution of Dry Milled Free Base Powder**

Dexrazoxane powder was dry milled and separated as in Example 4. Dissolution studies were conducted on powders with particle size ranging between >125-180µm, >90-125µm, >63-90µm, >45-63µm and >32-45µm. The dissolution studies mimicked the clinical procedures used in clinics and hospitals to dissolve drug products in infusion solutions prior to IV administration. The dissolution media were readily available infusion solutions usually stocked in hospitals: i.e. 0.9% sodium chloride USP and 5% Dextrose in Water, USP. The concentration of the drug powder was limited to 7.9 to 8.4 mg/mL to simulate 2000 mg dosage administrated in a 250ml infusion fluid. The dry powder was weighed in a 4mL glass vial and an adequate amount of infusion fluid was added onto it. The vial was shaken by hand at ambient temperature until the powder dissolved completely. The dissolution time was recorded. Results are shown in Tables 4 and 5.

**Example 6: Solubility, Stability and pH of Dexrazoxane Dry Milled Free Base Infusion Solutions**

HPLC solubility and stability analysis were performed using a Waters™ Alliance Model 2695 system consisting of a PDA UV detector model 2996, and pump model 2695 and an auto sampler. The data was acquired and analyzed on the Empower™ chromatography data system. The raw data are stored in a server.

Solution pH was measured using an Accumet model AB15 pH meter (Fisher Scientific International, Inc., NH) equipped with a Beckman PN 511063 glassed electrode.

Solubility studies were performed by shaking or agitating an excess dexrazoxane drug powder suspended in infusion solutions (5% Detrose, USP, 0.9% sodium chloride USP, Plasma-Lyte A USP, sodium lactate USP or Ringers lactate USP) for one hour at a constant temperature (25°C). Samples of the suspension were clarified by filtration through a 0.2µm Nylon (Pall) filters. The first filtered milliliter of solution was discarded and subsequent filtered solution was collected for assay by reversed-phase HPLC. All assayed solutions were quenched with 1mL of 0.1N...
hydrochloric acid to minimize the decomposition of the filtered solution. The quenched solutions were further adjusted with 0.5mL of 0.1 N sodium hydroxide solutions to pH 3.0 immediately before HPLC assay and diluted with 0.5mM disodium EDTA to within the linearity range of the assay method. Results appear in Table 3.

Stability was measured by dexrazoxane (ICRF-187) free base was dissolved in 5% Dextrose, 0.9% NaCl, Plasma-Lyte A, Na lactate or Ringers lactate at concentrations of 10 mg/mL or 4 mg/mL. Upon complete dissolution, the pH of each solution was measured and recorded. The solutions were stored at 25°C or 5°C. Stability samples were collected at 0, 2, 4, 6, 8, and 24 hours. Each sample was diluted with the HPLC mobile phase to a concentration of about 1.0 mg/mL prior to assay by HPLC. Stability was measured by calculating percentage (peak A + peak B + peak C) as in Example 3. Results appear in Figures 5-7.

Example 7: Preparation of Isolated Salt Crystals

A variety of methods were employed to crystallize the acid salts. The methods are described below and results are summarized in Table 2A:

Cooling Method

ICRF-187 freebase was dissolved in the desired crystallization solvent or mixture of solvents (Table 2A). These solvents were chosen such that the dexrazoxane freebase is soluble in the solvents but the salts formed from the reaction of the freebase and acids when the respective acids are added to the crystallization solvents are not soluble. Equimolar amount of the desired acid - i.e. sulfuric, maleic, methanesulfonic or hydrochloric acid —was added to the solvent to form a solution. The salt precipitated in the solution. The precipitates, which were typically not crystalline or were partially crystalline are collected through filtration on a filter and dried by vacuum. Methanol was then added to the dried powder and heated to 65°C to dissolve the entire salt. The solution then cooled in a controlled manner to 5°C overnight in RS-IO Chemblock Reaction Station (Barnstead International, Dubuque, IA). The crystals are then harvested on a filter and dried at ≤ 40°C.
Anti-Solvent Method

ICRF-187 freebase was dissolved in the desired crystallization solvent or mixture of solvents (Table 1). Equimolar amount of the acid - i.e. sulfuric, maleic, methanesulfonic or hydrochloric acid - was added to the solvent or mixtures of solvents to form a solution but the total solution volume is such that the salt formed in-situ remains dissolved in the solution or is soluble in the solution. While the solution was being gently stirred, an anti-solvent (Table 2A) was slowly added until the salt begins to precipitate from the solution. Additional anti-solvent is added until sufficient salt is formed. The crystals were then harvested on a filter and dried at ≤ 40°C.

Solvent Evaporation method

ICRF-187 freebase is dissolved in dioxane. Equimolar amount of the acid - i.e. sulfuric, maleic, methanesulfonic or hydrochloric acid - was added to the solvent or mixtures of solvents to form a solution. The salt precipitated in the solution. With gently stirring, minimum amount of water was added until the salt is completely dissolved. Nitrogen was introduced into the solution and the solvents were slowly evaporated until salt precipitation appeared. Additional anti-solvent is added until sufficient salt is formed. The crystals are then harvested on a filter and dried at ≤ 40°C.

Direct precipitation method

ICRF-187 freebase was dissolved in dioxane. Equimolar amount of the acid - i.e. sulfuric, maleic, methanesulfonic or hydrochloric acid - was added to the solvent or mixtures of solvents to form a solution. The salt precipitated in the solution. The crystals are then harvested on a filter and dried at < or = 40°C.

X-ray powder diffraction analyses

The XRPD analyses were performed using a Shimadzu XRD-6000 X-ray powder diffractometer using Cu Ka radiation. The instrument was equipped with a long fine focus X-ray tube. The tube voltage and amperage were set to 40 kV and 40
mA, respectively. The divergence and scattering slits were set at 1° and the receiving slit was set at 0.15 mm. Diffracted radiation was detected by a NaI scintillation detector. A θ2θ continuous scan at 37min (0.4 sec/0.02° step) from 2.5 to 40° 2θ was used. A silicon standard was analyzed to check the instrument alignment. Data were collected and analyzed using XRD-6000 v. 4.1. The peak lists were generated with the Shimadzu software using default peak picking parameters on smoothed, background-subtracted, Kα1-corrected traces.
What is claimed is:

1. A composition comprising a pharmaceutically acceptable salt of dexrazoxane (ICRF-187) in solution, wherein the composition has a concentration of at least 60mg/ml dexrazoxane.

2. The composition of claim 1 wherein the solution has a concentration of at least 80mg/mL dexrazoxane.

3. The composition of any of claims 1 or 2 wherein the salt is a bis, mono or sesqui-mono salt of dexrazoxane.

4. The composition of any of claims 1-3 wherein the salt is selected from a sulfate salt, a mesylate salt, a maleate salt, an oxalate salt, a malonate salt and a phosphate salt.

5. The composition of claim 4 wherein the salt is selected from a sulfate salt, a mesylate salt and a maleate salt.

6. The composition of any of claims 1, 2, 3 or 5 wherein the composition is an aqueous acid solution of the salt that is capable of being maintained at a temperature of 50°C without precipitation.

7. The composition of claim 6 wherein the composition is stable for at least 48 hours.

8. A lyophilized product of an acid salt of dexrazoxane, wherein the acid is selected from one of methanesulfonic, maleic, oxalic, phosphoric, and malonic acids.

9. The lyophilized product of claim 8 wherein the acid salt is prepared by dissolving at least 30 mg/ml dexrazoxane in the acid.
10. The lyophilized product of any of claims 8 or 9 wherein greater than 500 mg of
dexrazoxane is contained in a vial of no more than 50 ml.

11. The lyophilized product of any of claims 8-10 wherein the product is
reconstituted in a solution.

12. The lyophilized product of claim 11 wherein the concentration of dexrazoxane
in the reconstituted solution is diluted to a concentration ranging from about 4
mg/ml to 10 mg/ml.

13. The lyophilized product of any of claims 11 or 12 wherein the reconstitution
solution comprises at least one of sodium lactate, Ringers lactate, Plasma-Lyte
A or sodium chloride.

14. The lyophilized product of any of claims 11-13 wherein the compound is
stable for at least 6 hours, stability defined as having less than or equal to 3%
total decomposition product.

15. The lyophilized product of claim 14 wherein the compound is stable for at
least 8 hours.

16. The lyophilized product of any of claims 8-15 wherein said product comprises
a unit dosage of at least 2 grams of a sterile pharmaceutical composition of a
stable, lyophilized acid salt of dexrazoxane (ICRF-187) in a single vial of no
more than 50 ml.

17. A method of formulating a pharmaceutical composition comprising dissolving
at least 60 mg/ml of dexrazoxane (ICRF-187) in solution comprising an acid
of the salt.

18. The method of claim 17 wherein at least 80 mg/ml of dexrazoxane (ICRF-187)
is dissolved in the solution.
19. The method of any of claims 17 or 18 wherein the acid is selected from one of sulfuric, methanesulfonic, maleic, oxalic, phosphoric, and malonic acids.

20. The method of claim 19 wherein the acid is selected from one of sulfuric, methanesulfonic and maleic acids.

21. The method of any of claims 17-20 further comprising filling sterile vials with a volume of the solution of volume sufficient to formulate a 2 gram dose of dexrazoxane in each vial.

22. The method of claim 21 further comprising placing the vials, in a lyopliilizer to create a lyophilized product having less than 2% moisture content.

23. The method of claim 17 further comprising
   a) lyophilizing said solution comprising the acid of the salt to provide a lyophilizate;
   b) reconstituting the lyophilizate in a single vial in a solution having a concentration of at least 60mg/mL; and
   c) diluting the reconstituted solution in an intravenous to a final concentration of 4mg/mL to 10mg/mL for administration to the human.

24. A composition comprising a non-lyophilized sterile dexrazoxane free base powder, wherein the powder has a mean particle size of less than 100 μm.

25. The composition of claim 24 wherein the powder has a mean particle size of less than 50 μm.

26. The composition of any of claims 24 or 25 wherein the powder is dissolved in an infusion solution.

27. The composition of claim 26 wherein the infusion solution has a pH of at least 5.5.
28. The composition of any of claims 24-27 wherein at least 80% of the particle size distribution is between 32 and 45 µm.

29. The composition of any of claims 24-28 wherein the powder is amorphous.

30. The composition of any of claims 24-28 wherein the powder is crystalline.


32. The composition of any of claims 24-31 wherein the powder is capable of being dissolved in an infusion solution in under five minutes.

33. The composition of any of claims 24-32 wherein the powder is capable of being fully dissolved in an infusion solution prior to decomposition of dexrazoxane.

34. The composition of any of claims 24-33, wherein the dexrazoxane is packaged in a receptacle suitable for the addition of a buffer fluid to the powder.

35. A packaged dexrazoxane product comprising a non-lyophilized sterile dexrazoxane free base powder having a mean particle size of less than 100 µm, wherein the dexrazoxane is packaged in a receptacle suitable for the addition of a buffer fluid to the powder.

36. The packaged dexrazoxane product of claim 35 wherein the receptacle is configured for a sterile transfer of the powder to a bag or bottle containing the buffer.
37. The packaged dexrazoxane product of claim 36 wherein the receptacle is a vial attached to an infusion bag.

38. The packaged dexrazoxane product of claim 36 wherein the receptacle is a dockable vial.

39. The packaged dexrazoxane product of claim 36 wherein the receptacle is an IV infusion bottle or bag.

40. A method of formulating a pharmaceutical composition comprising dry milling dexrazoxane free base or acid salt crystals to form a dry milled powder.

41. The method of claim 40 further comprising sieving the dry milled powder.

42. The method of claim 40 wherein dry milling the free base or acid salt crystals comprises using a ball mill or turbula mixer.

43. The method of any of claims 40-42 wherein dexrazoxane free base crystals are dry milled.

44. A method of formulating dexrazoxane for IV administration comprising dissolving dexrazoxane free base in an IV solution to a concentration of 10 mg/ml in less than 5 minutes.

45. The method of claim 44, wherein the dexrazoxane is administered to a patient prior to administration of doxorubicin.

46. The method of any of claims 44 or 45 wherein the free base is stable for at least 6 hours in the solution.
47. A method of treating cardiotoxicity associated with the administration of an anthracycline comprising administering the compositions of any of claims 1-7 or 24-33 to a patient.

48. The method of claim 47, wherein said anthracycline is doxorubicin.

49. The method of any of claims 47 or 48, wherein administration of the composition is substantially without pain associated with the administration of low-pH and/or hydrochloric acid containing solutions.

50. The method of any of claims 47-49, wherein the composition is administered to the subject prior to administration of the anthracycline.

51. A method of treating cardiotoxicity associated with the administration of an anthracycline comprising administering the lyophilized product of any of claims 8-16 to a patient.
Comparison of mesylate and sulfate stability with current lyo product after dilution with infusion fluid to 10 mg/ml; samples stored for 8 hours at 25 C after dilution

FIG. 1
Comparison of mesylate and sulfate stability with current lyo product after dilution with infusion fluid to 4 mg/ml; samples stored for 8 hours at 25°C after dilution.
Stability of maleate, sulfate and mesylate salt forms vs. current lyo product (HCl salt) at 25°C

**FIG. 3**
Stability of maleate, sulfate and mesylate salt forms vs. current lyo product (HCl salt) at 5C

FIG. 4
Stability of maleate, sulfate and mesylate salt forms vs. current Lyo product (HCl salt) at 5°C

Product Spec for A+B+C in Drug Product = 2.5%

- Maleate salt 30 mg/mL
- Sulfate salt 30 mg/mL
- Mesylate salt 30 mg/mL
- Current Lyo product 20 mg/mL

FIG. 4A
Stability of dexrazoxane free base at 25°C after dilution with infusion fluid to 10 mg/ml

FIG. 5
Stability of dexrazoxane free base at 25C after dilution with infusion fluid to 4 mg/ml

% A + B + C

0.00 0.50 1.00 1.50 2.00 2.50 3.00 3.50 4.00

4 6 8 24

Time (hours)

5.63 6.20 6.51 8.83 1.00

5% dextrose (pH 5.85)
0.9% NaCl (pH 5.69)
Plasmalyte A (pH 6.88)
Na lactate (pH 6.49)
Ringer lactate (pH 6.25)

FIG. 6
Stability of dexrazoxane free base at 5C after dilution with infusion fluid to 4 mg/ml

FIG. 7
Add API to Sterile WFI in a Jacketed tank and Heat

Filter while hot through 0.22μm filters into transport vessel

Crystallize Sterile API in Class 100 room

Collect crystals in filter and tray-dry in Class 100 room

Mill crystals to smaller size

Fill into Sterile Vials

Stopper Vials

QC Testing

FIG. 8