TAZOBACTAM AND CEFTOLOZANE ANTIBIOTIC COMPOSITIONS

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ABSTRACT

This disclosure provides pharmaceutical compositions comprising ceftolozane, pharmaceutical compositions comprising ceftolozane and tazobactam, methods of making those compositions, and related methods and uses of these compositions.
Fig. 1

Atmospheric pressure

100 to 150 mTorr

-40°C

-20°C

30°C

NMT 10 mTorr

Chamber pressure

Shelf temperature
Fig. 2

CXA-101 drug substance

Water for Injection

Citric acid, anhydrous

Sodium bicarbonate

Sodium Chloride

Maltose

Weighing

Weighing

Weighing

Weighing

Weighing

Weighing

Weighing

Dissolution

Suspension

Dissolution

Dissolution

Nitrogen purge

5% Sodium bicarbonate

Water for Injection

pH adjustment

Weight adjustment

Prefiltration

Aseptic filtration

Nitrogen

Aseptic filtration

Sterilization

Sterilization

Sterilization

Vial

Stopper

Flip-off Cap

Filling and half-stopping

Lyophilization

Stoppering and crimping (Grade A)

Visual inspection

CXA-101 lyophilized product

Aseptic room (Grade B)
A. DISSOLUTION
1) 81 kg WFI
2) 196 g Anhydrous citric acid
4) 11.4 kg CXA-101 (9.462 kg potency)
5) 5.55 Kg L-arginine base
7) 4.5 kg NaCl
8) pH adjustment (range 6.0 - 7.0 with L-arginine or citric acid only if necessary
9) Add WFI to total 124.4 kg while mixing

DISSOLUTION REACTOR

→ 3) Temperature check at 5°C-10°C while stirring
→ 6) Complete dissolution check pH = 6.5-7.0

→ 10) Samples withdrawn for testing

B. STERILE FILTRATION
11) Sterile filtration and load into lyophilizer
Filter 0.45 µm porosity → Class 100,000 room
Filter 0.22 µm porosity → Class 1000 room
Filter 0.22 µm porosity → Class 100 room

→ 12) WFI line wash
→ 13) Washing solution put through steps 12 to 14

C. BULK LYOPHILIZATION
14) Sterile filtered washing solution placed into lyophilizer on a separate shelf
15) Start lyophilization

CRIOFARMA LYOPHILIZER

→ 16) Temperature monitoring of product shelf 20°C ± 5°C

D. PACKAGING INTO STERBAGS®
17) Milling of powder
GRINDER → Class 1000 room

→ 18) Sieving of powder
SIEVE → Class 1000 room

→ 19) Blending of powder (30 min)
BLENDER → Class 1000 room

→ 20) Discharge into Sterbags®
17.4-KG STERBAGS® (Yield 19.1 kg ± 10%)

Class 100 room

Fig. 3
Bulk drug product of CXA-101/tazobactam for injection packaged in Sterbag®

Outer bag surface sterilized in UV box
Outer bag removed in LAF hood

Middle bag surface sterilized in UV box
Sterile middle bag removed under LAF

Sterile inner bag attached to filling machine powder hopper
Drug product transferred by gravity under LAF

Bulk drug product filled into sterile vials by filling machine under a nitrogen blanket

Vials stoppered with sterilized rubber stoppers

Vials sealed with sterilized caps

Outside of vials washed

All vials visually inspected

Vials labeled and packaged

Fig. 4
Fig. 5
The Purity of Ceftolozane, 60°C

**Fig. 6**
Fig. 7

Peak 1, 60°C

- □ 190.0 mg NaCl per 1000 mg of ceftolozane
- ★ 125.0 mg NaCl per 1000 mg of ceftolozane
- ▲ 75.0 mg NaCl per 1000 mg of ceftolozane
- × 50.0 mg NaCl per 1000 mg of ceftolozane
- ★★ 481.0 mg NaCl per 1000 mg of ceftolozane

Days

Peak Area
Peak 7, 60°C

Fig. 9
The Purity of Ceftolozane, 60°C

Fig. 10
Peak 1, 60°C

**Fig. 11**
RRT 0.43 + Peak 3, 60°C

Fig. 12
Peak 7, 60°C

Fig. 13
Non-sterile APIs

Tazobactam

CXA-101

Dissolution, Sterile Filtration

Tray Co-lyophilization

Vial Fill-Finish

Fig. 14
Fig. 15
TAZOBACTAM AND CEFTOLOZANE ANTIBOTIC COMPOSITIONS

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation of U.S. application Ser. No. 14/214,367, filed Mar. 14, 2014, which claims priority to U.S. Provisional Application No. 61/792,092, filed Mar. 15, 2013, and U.S. Provisional Application No. 61/793,007, filed Mar. 15, 2013. The contents of these applications are incorporated hereby by reference in their entireties.

TECHNICAL FIELD

[0002] This disclosure relates to pharmaceutical compositions comprising ceftolozane, pharmaceutical compositions comprising tazobactam and ceftolozane, methods of making those compositions, and related methods and uses thereof.

BACKGROUND

[0003] The cephalosporin (6R,7R)-3-[(5-amino-4-[(2-a-minothiolethoxy)iminio]carboxamido)carbonyl]amino]-1-methyl-1H-pyrazol-2-i um-2-yl]methyl]-7-[(6Z)-2-(5-amino-1,2,4-thiazolidine-3-yl)-2-[(1-carboxy-1-methylthiolethox)iminio]acetyl]amino]-8-oxo-5-throw-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylate (also referred to as ceftolozane, “CAX-101”) or (6R,7R)-3-[5-Amino-4-[(2-aminothiolethoxy)iminio]carboxamido]carbonyl]amino]-1-methyl-1H-pyrazol-2-ium-2-ylmethyl]-7-[(5-amino-1,2,4-thiazolidine-3-yl)-2-[(1-carboxy-1-methylthiolethox)iminio]acetamido]-3-cephem-4-carboxylic acid) is an antibacterial agent. The antibacterial activity of ceftolozane is believed to result from its interaction with penicillin binding proteins (PBPs) to inhibit the biosynthesis of the bacterial cell wall which acts to stop bacterial replication. Cefeprome can be combined (e.g., mixed) with a 2-lactamase inhibitory (“BLI”), such as tazobactam. Tazobactam is a BLI against Class A and some Class C bluntamases, with well-established in vitro and in vivo efficacy in combination with active 2-lactam antibiotics.

[0004] Antibiotic pharmaceutical compositions can include a beta-lactam compound having antibacterial properties (i.e., an antibiotic compound possessing one or more beta-lactam moieties) and a BLI, such as tazobactam. Beta-lactam compounds can be formulated with and/or administered in combination with, beta-lactamase inhibiting compounds (e.g., tazobactam and salts thereof) in order to mitigate the effects of bacterial beta-lactamases. For example, the combination of ceftolozane, and tazobactam in a 2:1 weight ratio is an antibiotic pharmaceutical composition (“CAX-201”) formulated for parenteral administration. CAX-201 displays potent antibacterial activity in vitro against common Gram-negative and selected Gram-positive organisms. CAX-201 is a broad-spectrum antibiotic with in vitro activity against Enterobacteriaceae including strains expressing extended spectrum 2-lactamases-resistant (MIC_{90} = 1 μg/mL), as well as Pseudomonas aeruginosa (P. aeruginosa) including multidrug resistant strains (MIC_{90} = 2 μg/mL). CAX-201 is a combination antibacterial with activity against many Gram-negative pathogens known to cause intrapulmonary infections, including nosocomial pneumonia caused by P. aeruginosa.

SUMMARY OF THE INVENTION

[0005] Provided herein are pharmaceutical compositions comprising ceftolozane, methods of making the compositions, and pharmaceutical compositions prepared using ceftolozane. Also provided herein are pharmaceutical compositions comprising ceftolozane and tazobactam, methods of making those compositions, and pharmaceutical compositions prepared using ceftolozane and tazobactam. Methods of making and related uses of these combinations are also provided.

[0006] In one aspect, provided herein is pharmaceutical composition comprising sodium chloride and 7β-[(6Z)-2-(5-amino-1,2,4-thiazolidine-3-yl)-2-(1-carboxy-1-methylthiolethox)iminio]acetamido]-3-[3-amino-4-[(2-aminothiolethoxy)iminio]carboxamido]-2-methyl-1-pyrrozolo]-methyl-3-cephem-4-carboxylate at a weight ratio of 476:1000 to 500:1000.

[0007] In one particular embodiment, the sodium chloride and 7β-[(6Z)-2-(5-amino-1,2,4-thiazolidine-3-yl)-2-(1-carboxy-1-methylthiolethox)iminio]acetamido]-3-[3-amino-4-[(2-aminothiolethoxy)iminio]carboxamido]-2-methyl-1-pyrrozolo]-methyl-3-cephem-4-carboxylate are at a weight ratio of 476:1000.

[0008] In another particular embodiment, the sodium chloride and 7β-[(6Z)-2-(5-amino-1,2,4-thiazolidine-3-yl)-2-(1-carboxy-1-methylthiolethox)iminio]acetamido]-3-[3-amino-4-[(2-aminothiolethoxy)iminio]carboxamido]-2-methyl-1-pyrrozolo]-methyl-3-cephem-4-carboxylate are at a weight ratio of 480:1000.

[0009] In a further embodiment, the sodium chloride and 7β-[(6Z)-2-(5-amino-1,2,4-thiazolidine-3-yl)-2-(1-carboxy-1-methylthiolethox)iminio]acetamido]-3-[3-amino-4-[(2-aminothiolethoxy)iminio]carboxamido]-2-methyl-1-pyrrozolo]-methyl-3-cephem-4-carboxylate are at a weight ratio of 481:1000 to 500:1000.

[0010] In one particular embodiment, the sodium chloride and 7β-[(6Z)-2-(5-amino-1,2,4-thiazolidine-3-yl)-2-(1-carboxy-1-methylthiolethox)iminio]acetamido]-3-[3-amino-4-[(2-aminothiolethoxy)iminio]carboxamido]-2-methyl-1-pyrrozolo]-methyl-3-cephem-4-carboxylate are at a weight ratio of 481:1000.

[0011] In yet another particular embodiment, the sodium chloride and 7β-[(6Z)-2-(5-amino-1,2,4-thiazolidine-3-yl)-2-(1-carboxy-1-methylthiolethox)iminio]acetamido]-3-[3-amino-4-[(2-aminothiolethoxy)iminio]carboxamido]-2-methyl-1-pyrrozolo]-methyl-3-cephem-4-carboxylate are at a weight ratio of 485:1000.

[0012] In certain embodiments, the pharmaceutical compositions comprise less than 4% by weight of water.

[0013] In another aspect, provided herein is a pharmaceutical composition comprising sodium chloride and 7β-[(6Z)-2-(5-amino-1,2,4-thiazolidine-3-yl)-2-(1-carboxy-1-methylthiolethox)iminio]acetamido]-3-[3-amino-4-[(2-aminothiolethoxy)iminio]carboxamido]-2-methyl-1-pyrrozolo]-methyl-3-cephem-4-carboxylate at a molar ratio of 8.14:1 to 8.56:1.

[0014] In one embodiment, provided herein is a pharmaceutical composition comprising sodium chloride and 7β-[(6Z)-2-(5-amino-1,2,4-thiazolidine-3-yl)-2-(1-carboxy-1-methylthiolethox)iminio]acetamido]-3-[3-amino-4-[(2-aminothiolethoxy)iminio]carboxamido]-2-methyl-1-pyrrozolo]-methyl-3-cephem-4-carboxylate at a molar ratio of 8.23:1 to 8.56:1.

[0015] In another aspect, provided herein is a vial containing an antibiotic composition for treating an infection, wherein the antibiotic composition comprises sodium chlo-
ride and 7β-[(Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(1-carboxy-1-methylethoxyimino)acetamido]-3-3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-pyrazolyl]methyl-3-cephem-4-carboxylate at a weight ratio of 476:1000 to 500:1000.

[0016] In one particular embodiment of the vial, the antibiotic composition comprises sodium chloride and 7β-[(Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(1-carboxy-1-methylethoxyimino)acetamido]-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-pyrazolyl]methyl-3-cephem-4-carboxylate at a weight ratio of 476:1000.

[0017] In another particular embodiment of the vial, the antibiotic composition comprises sodium chloride and 7β-[(Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(1-carboxy-1-methylethoxyimino)acetamido]-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-pyrazolyl]methyl-3-cephem-4-carboxylate at a weight ratio of 480:1000.

[0018] In a further embodiment of the vial, the antibiotic composition comprises sodium chloride and 7β-[(Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(1-carboxy-1-methylethoxyimino)acetamido]-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-pyrazolyl]methyl-3-cephem-4-carboxylate at a weight ratio of 481:1000 to 500:1000.

[0019] In one particular embodiment of the vial, the antibiotic composition comprises sodium chloride and 7β-[(Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(1-carboxy-1-methylethoxyimino)acetamido]-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-pyrazolyl]methyl-3-cephem-4-carboxylate at a weight ratio of 481:1000.

[0020] In yet another particular embodiment of the vial, the antibiotic composition comprises sodium chloride and 7β-[(Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(1-carboxy-1-methylethoxyimino)acetamido]-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-pyrazolyl]methyl-3-cephem-4-carboxylate at a weight ratio of 485:1000.

[0021] In certain embodiments of the vials described above, the antibiotic compositions comprise less than 4% by weight of water.

[0022] In other embodiments of the vials, the infections are caused by bacteria selected from the group consisting of Staphylococcus aureus, Escherichia coli, Acinetobacter baumannii, Haemophilus influenzae, Klebsiella pneumoniae, and Pseudomonas aeruginosa.

[0023] In other embodiments of the vials, the infections are selected from the group consisting of nosocomial pneumonia, complicated intra-abdominal infections and complicated urinary tract infections.

[0024] In still another aspect, provided herein is a method of preparing a composition comprising sodium chloride and 7β-[(Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(1-carboxy-1-methylethoxyimino)acetamido]-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-pyrazolyl]methyl-3-cephem-4-carboxylate, wherein the method comprises combining sodium chloride with 7β-[(Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(1-carboxy-1-methylethoxyimino)acetamido]-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-

pyrazolyl]methyl-3-cephem-4-carboxylate at a weight ratio of 476:1000 to 500:1000 to form a mixture, followed by lyophilization of the mixture.

[0025] In one embodiment, the method comprises combining sodium chloride with 7β-[(Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(1-carboxy-1-methylethoxyimino)acetamido]-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-

pyrazolyl]methyl-3-cephem-4-carboxylate at a weight ratio of 481:1000 to 500:1000 to form a mixture, followed by lyophilization of the mixture.

[0026] In a further aspect, provided herein is a method of preparing a composition comprising sodium chloride and 7β-[(Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(1-carboxy-1-methylethoxyimino)acetamido]-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-

pyrazolyl]methyl-3-cephem-4-carboxylate, wherein the method comprises combining sodium chloride with 7β-[(Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(1-carboxy-1-methylethoxyimino)acetamido]-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-

pyrazolyl]methyl-3-cephem-4-carboxylate at a molar ratio of 8.14:1 to 8.56:1 to form a mixture, followed by lyophilization of the mixture.

[0027] In one embodiment, the method comprises combining sodium chloride with 7β-[(Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(1-carboxy-1-methylethoxyimino)acetamido]-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-

pyrazolyl]methyl-3-cephem-4-carboxylate at a molar ratio of 8.23:1 to 8.56:1 to form a mixture, followed by lyophilization of the mixture.

[0028] In another aspect, provided herein is a composition formed by any one of the methods provided herein.

[0029] In yet another aspect, provided herein is a method for the treatment of a bacterial infection in a mammal, wherein the method comprises administering to said mammal a therapeutically effective amount of a pharmaceutical composition provided herein.

[0030] In one embodiment of the treatment method, the bacterial infection is caused by bacteria selected from the group consisting of Staphylococcus aureus, Escherichia coli, Acinetobacter baumannii, Haemophilus influenzae, Klebsiella pneumoniae, and Pseudomonas aeruginosa.

[0031] In one embodiment of the treatment method, the bacterial infection is selected from the group consisting of nosocomial pneumonia, complicated intra-abdominal infections and complicated urinary tract infections.

**BRIEF DESCRIPTION OF THE FIGURES**

[0032] FIG. 1 is a graph showing the lyophilization program used in the manufacturing of the monoprotect for injection.

[0033] FIG. 2 is a flowchart showing the manufacturing process for a CXA-101 composition for injection.

[0034] FIG. 3 is a flowchart showing the manufacturing steps for a pharmaceutical composition comprising cefotolozane and sodium chloride.
FIG. 4 is a flowchart showing the manufacturing process for preparing a CXA-201 composition comprising ceftolozane (referred to as CXA-101), tazobactam, and sodium chloride.

FIG. 5 is a reference HPLC chromatogram showing the peaks of ceftolozane (also referred to as CXA-101) and related impurities.

FIG. 6 is a plot of the data points from Table 5, showing the purity of the ceftolozane in CXA-101 compositions at 60°C on day 0, day 1, day 3, and day 7, as measured by the HPLC method described in Example 4, wherein the CXA-101 compositions comprise ceftolozane and sodium chloride. The amount of the sodium chloride in the CXA-101 compositions is 481.0 mg sodium chloride per 1000 mg of ceftolozane (* marks), 190.0 mg sodium chloride per 1000 mg of ceftolozane (filled squares), 125.0 mg sodium chloride per 1000 mg of ceftolozane (filled triangles), 75.0 mg sodium chloride per 1000 mg of ceftolozane (filled diamonds), and 50.0 mg sodium chloride per 1000 mg of ceftolozane (X marks).

FIG. 7 is a plot of the data points from Table 6, showing the peak area of the impurity peak 1 in CXA-101 compositions at 60°C on day 0, day 1, day 3, and day 7, as measured by the HPLC method described in Example 4, wherein the CXA-101 compositions comprise ceftolozane and sodium chloride. The amount of the sodium chloride in the CXA-101 compositions is 481.0 mg sodium chloride per 1000 mg of ceftolozane (* marks), 190.0 mg sodium chloride per 1000 mg of ceftolozane (filled squares), 125.0 mg sodium chloride per 1000 mg of ceftolozane (filled diamonds), 75.0 mg sodium chloride per 1000 mg of ceftolozane (filled triangles), and 50.0 mg sodium chloride per 1000 mg of ceftolozane (X marks).

FIG. 8 is a plot of the data points from Table 7, showing the total peak area of the impurity with a RRT of 0.43 and the impurity peak 3 in CXA-101 compositions at 60°C on day 0, day 1, day 3, and day 7, as measured by the HPLC method described in Example 4, wherein the CXA-101 compositions comprise ceftolozane and sodium chloride. The amount of the sodium chloride in the CXA-101 compositions is 481.0 mg sodium chloride per 1000 mg of ceftolozane (* marks), 190.0 mg sodium chloride per 1000 mg of ceftolozane (filled squares), 125.0 mg sodium chloride per 1000 mg of ceftolozane (filled diamonds), 75.0 mg sodium chloride per 1000 mg of ceftolozane (filled triangles), and 50.0 mg sodium chloride per 1000 mg of ceftolozane (X marks).

FIG. 9 is a plot of the data points from Table 8, showing the peak area of the impurity peak 7 in CXA-101 compositions at 60°C on day 0, day 1, day 3, and day 7, as measured by the HPLC method described in Example 4, wherein the CXA-101 compositions comprise ceftolozane and sodium chloride. The amount of the sodium chloride in the CXA-101 compositions is 481.0 mg sodium chloride per 1000 mg of ceftolozane (* marks), 190.0 mg sodium chloride per 1000 mg of ceftolozane (filled squares), 125.0 mg sodium chloride per 1000 mg of ceftolozane (filled diamonds), 75.0 mg sodium chloride per 1000 mg of ceftolozane (filled triangles), and 50.0 mg sodium chloride per 1000 mg of ceftolozane (X marks).

FIG. 10 is a plot of the data points from Table 10, showing the purity of ceftolozane in CXA-201 compositions at 60°C on day 0, day 1, day 3, and day 7, as measured by the HPLC method described in Example 4, wherein the CXA-201 compositions comprise ceftolozane, tazobactam, and sodium chloride. The amount of the sodium chloride in the CXA-201 compositions is 481.0 mg sodium chloride per 1000 mg of ceftolozane (X marks), 125.0 mg sodium chloride per 1000 mg of ceftolozane (filled triangles), 75.0 mg sodium chloride per 1000 mg of ceftolozane (filled squares), and 50.0 mg sodium chloride per 1000 mg of ceftolozane (filled triangles).

FIG. 11 is a plot of the data points from Table 11, showing the peak area of the impurity peak 1 in CXA-201 compositions at 60°C on day 0, day 1, day 3, and day 7, as measured by the HPLC method described in Example 4, wherein the CXA-201 compositions comprise ceftolozane, tazobactam, and sodium chloride. The amount of the sodium chloride in the CXA-201 compositions is 481.0 mg sodium chloride per 1000 mg of ceftolozane (X marks), 125.0 mg sodium chloride per 1000 mg of ceftolozane (filled triangles), 75.0 mg sodium chloride per 1000 mg of ceftolozane (filled squares), and 50.0 mg sodium chloride per 1000 mg of ceftolozane (filled triangles).

FIG. 12 is a plot of the data points from Table 12, showing the total peak area of the impurity with a RRT of 0.43 and the impurity peak 3 in CXA-201 compositions at 60°C on day 0, day 1, day 3, and day 7, as measured by the HPLC method described in Example 4, wherein the CXA-201 compositions comprise ceftolozane, tazobactam, and sodium chloride. The amount of the sodium chloride in the CXA-201 compositions is 481.0 mg sodium chloride per 1000 mg of ceftolozane (X marks), 125.0 mg sodium chloride per 1000 mg of ceftolozane (filled triangles), 75.0 mg sodium chloride per 1000 mg of ceftolozane (filled squares), and 50.0 mg sodium chloride per 1000 mg of ceftolozane (filled triangles).

FIG. 13 is a plot of the data points from Table 13, showing the peak area of the impurity peak 7 in CXA-201 compositions at 60°C on day 0, day 1, day 3, and day 7, as measured by the HPLC method described in Example 4, wherein the CXA-201 compositions comprise ceftolozane, tazobactam, and sodium chloride. The amount of the sodium chloride in the CXA-201 compositions is 481.0 mg sodium chloride per 1000 mg of ceftolozane (X marks), 125.0 mg sodium chloride per 1000 mg of ceftolozane (filled squares), and 50.0 mg sodium chloride per 1000 mg of ceftolozane (filled triangles).

FIG. 14 is a manufacturing flowchart showing CXA-201 development using a co-lyophilization process, as described herein.

FIG. 15 is the formula of the impurity RRT 1.22, which has been identified to be a degradation product formed by a reaction between ceftolozane and formylacetic acid, a

**DETAILED DESCRIPTION OF THE INVENTION**

[0047] Pharmaceutical compositions comprising one or more drug substances or excipients can be prepared in a variety of ways, including, for example, blending and lyophilization (also known as “co-lyophilization”). As is known to those skilled in the art, lyophilization is a process of freeze-drying in which water is sublimed from a frozen solution of one or more solutes. Specific methods of lyophilization are described in Remington’s Pharmaceutical Sciences, Chapter 84, page 1565, Eighteenth Edition, A. R. Gennaro, (Mack Publishing Co., Easton, Pa., 1990).

[0048] The formulation of pharmaceutical compositions can be selected to minimize decomposition of the constituent drug substances and to produce a composition that is stable under a variety of storage conditions.

[0049] Surprisingly, pharmaceutical compositions comprising ceftolozane and 125 to 1000 mg sodium chloride per 1000 mg of ceftolozane have been observed to exhibit better chemical stability over the course of time and/or in the presence of heat, and less impurities than those pharmaceutical compositions comprising ceftolozane and less sodium chloride (i.e., less than 125 mg sodium chloride per 1000 mg of ceftolozane). In particular embodiments described herein provided herein is a pharmaceutical composition comprising ceftolozane and 125 to 1000 mg sodium chloride per 1000 mg of ceftolozane. In another aspect, the invention is a pharmaceutical composition comprising ceftolozane and tazobactam, further comprising 125 to 1000 mg sodium chloride per 1000 mg of ceftolozane. In certain embodiments of both of these aspects, the pharmaceutical composition comprises 125 to 500 mg sodium chloride per 1000 mg of ceftolozane.

[0051] As used herein, “125 to 1000 mg sodium chloride per 1000 mg of ceftolozane” refers to a ratio of sodium chloride to ceftolozane. For example, “125 to 1000 mg sodium chloride per 1000 mg of ceftolozane” includes, for example, 62.5 to 500 mg sodium chloride per 500 mg of ceftolozane, as well as, for example, 25 to 200 mg sodium chloride per 200 mg ceftolozane, etc.

[0052] In addition, surprisingly, pharmaceutical compositions comprising ceftolozane and tazobactam prepared by blending these two compounds, wherein the ceftolozane and tazobactam are individually lyophilized prior to blending, have been observed to exhibit beneficial properties, including reduced levels of impurities. In a particular embodiment described herein (see, e.g., Example 3), the pharmaceutical composition prepared by blending ceftolozane and tazobactam, wherein the ceftolozane and tazobactam were individually lyophilized prior to blending, lead to a much lower concentration of the following degradation product:

(see, e.g., Example 5), the pharmaceutical compositions comprising ceftolozane and 125 to 500 mg sodium chloride per 1000 mg of ceftolozane have been found to be more stable than the compositions comprising ceftolozane and less than 125 mg sodium chloride per 1000 mg of ceftolozane.

[0050] It has also been observed that pharmaceutical compositions comprising ceftolozane, tazobactam, and 125 to 1000 mg sodium chloride per gram of ceftolozane exhibit better chemical stability and less impurities than those pharmaceutical compositions comprising ceftolozane and tazobactam, but less sodium chloride. In particular embodiments described herein (see, e.g., Example 5), the pharmaceutical compositions comprising ceftolozane, tazobactam, and 125 to 500 mg sodium chloride per 1000 mg of ceftolozane have been found to be more stable than the compositions comprising ceftolozane, tazobactam, and less than 125 mg sodium chloride per gram of ceftolozane. Accordingly, in one aspect,

[0053] This degradation product, having a Relative Retention Time (RRT) of 1.22 (relative to ceftolozane using the HPLC method described in Example 4), which is also referred to as “the Impurity RRT 1.22”, has been identified to be a degradation product formed by a reaction between ceftolozane and formylacetic acid, a degradation product of tazobactam as illustrated in Marunaka et al. (Chem. Pharm. Bull. 1988, Vol. 36 (11), pp. 4478-4487).

[0054] In contrast, a much greater amount of this impurity was found in compositions of ceftolozane and tazobactam, wherein the compositions were formed through co-lyophilization, i.e., the ceftolozane and tazobactam were combined and lyophilized together, as opposed to being individually lyophilized (see, e.g., Example 7).

[0055] Accordingly, in one aspect, provided herein is a pharmaceutical composition comprising ceftolozane and tazobactam, wherein the composition has less than 0.1% by weight of the following compound:
In an embodiment, the pharmaceutical composition has less than 0.05% by weight of the following compound:

Ceftriaxone

The compound 5-amino-4-[(2-aminoethyl)carbamoyl]amino]-2-[[6R,7R]-7-[(2Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-[(1-carboxy-1-methylethoxy)imino]acetyl]amino]-2-carboxy-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-en-3-yl[methyl]-1-methyl-H-pyrazolium monosulfate (also known also as ceftriaxone sulfate, FR264205, “CXA-101”) is a cephalosporin compound (shown below), the synthesis of which is described in U.S. Pat. No. 7,129,232, wherein the compound is also named 7R-[Z]-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(1-carboxy-1-methylethoxyimino)acetamido]-3-[3-amino-4-[3-(2-aminoethyl)ureidol]-2-methyl-1-pyrazolol][methyl]-3-cephem-4-carboxylate. As used herein, the term “ceftolozane” may also refer to “ceftolozane sulfate”.

Pharmaceutical Compositions

The term “pharmaceutical composition” includes preparations suitable for administration to mammals, e.g., humans. When the compounds of the present invention are administered as pharmaceuticals to mammals, e.g., humans, they can be given per se or as a pharmaceutical composition containing, for example, 0.1% to 99.9% (more preferably, 0.5 to 90%) of active ingredient in combination with a pharmaceutically acceptable carrier.

The pharmaceutical compositions described herein can be formulated to have any concentration desired (i.e., any concentration of tazobactam, or a hydride or solvate thereof, and any concentration of ceftolozane). In some embodiments, the composition is formulated such that it comprises at least a therapeutically effective amount of both compounds (i.e., a therapeutically effective amount of the combination of tazobactam, or a hydrate or solvate thereof, and ceftolozane).

Pharmaceutical compositions include those suitable for parenteral (including intravenous) administration, although the most suitable route will depend on the nature and severity of the condition being treated. The compositions may be conveniently presented in unit dosage form, and prepared by any of the methods well known in the art of pharmacy.

Pharmaceutical compositions may additionally comprise excipients, stabilizers, pH adjusting additives (e.g., buffers) and the like. Non-limiting examples of these additives include sodium chloride, citric acid and L-arginine. For example, the use of sodium chloride results in greater stabil-
ity; L-arginine is used to adjust pH and to increase the solubility of ceftolozane; and citric acid is used to prevent discoloration of the product, due to its ability to chelate metal ions.

[0062] The pharmaceutical compositions disclosed herein can be prepared via blending. As used herein, blending refers to a process comprising physically combining ceftolozane and tazobactam, wherein each of ceftolozane and tazobactam have been individually lyophilized (i.e., lyophilized in the absence of one another) prior to being combined.

[0063] In a particular embodiment, the pharmaceutical compositions described herein are formulated for parenteral administration. In another particular embodiment, the pharmaceutical compositions described herein are formulated for administration by intravenous injection or infusion.

[0064] In one aspect, provided herein is a pharmaceutical composition comprising tazobactam and ceftolozane.

[0065] In another aspect, provided herein are pharmaceutical compositions prepared according to the following methods.

[0066] In one embodiment, provided herein is a pharmaceutical composition comprising sodium chloride and 7\(\beta\)-(\(\text{Z}\)-2-(5-amino-1,2,4-thiadiazole-3-yl)-2-(1-carboxy-1-methylthioxyiminio)acetamido)-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-pyrazolyl]methyl-3-cephem-4-carboxylate at a weight ratio of 476:1000 to 500:1000, or at a molar ratio of 8.14:1 to 8.56:1.

[0067] In another embodiment, provided herein is a pharmaceutical composition comprising sodium chloride and 7\(\beta\)-(\(\text{Z}\)-2-(5-amino-1,2,4-thiadiazole-3-yl)-2-(1-carboxy-1-methylthioxyiminio)acetamido)-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-pyrazolyl]methyl-3-cephem-4-carboxylate at a weight ratio of 481:1000 to 500:1000, or at a molar ratio of 8.23:1 to 8.56:1.

[0068] In certain particular embodiments, provided herein are pharmaceutical compositions comprising sodium chloride and 7\(\beta\)-(\(\text{Z}\)-2-(5-amino-1,2,4-thiadiazole-3-yl)-2-(1-carboxy-1-methylthioxyiminio)acetamido)-3-[3-amino-4-[3-(2-aminoethyl)ureido]-2-methyl-1-pyrazolyl]methyl-3-cephem-4-carboxylate at a weight ratio of 476:1000, or 480:1000, or 481:1000, or 485:1000.

[0069] In certain embodiments, the pharmaceutical compositions further comprise tazobactam sodium at a quantity equivalent of 500 mg of tazobactam free acid in a lyophilized powder form per 1000 mg of ceftolozane (amnhydrous, free base equivalent).

[0070] In certain embodiments, the pharmaceutical compositions comprise less than 4% by weight of water.

[0071] In other embodiments, the pharmaceutical compositions are reconstituted with sterile saline and/or sterile water for injection.

[0072] In a further embodiment, provided herein is a vial containing one of the pharmaceutical compositions described above for treating an infection. In certain embodiments, the infections are caused by bacteria selected from the group consisting of: Staphylococcus aureus, Escherichia coli, Acinetobacter baumanii, Haemophilus influenzae, Klebsiella pneumoniae, and Pseudomonas aeruginosa. In other embodiments, the infections are selected from the group consisting of nosocomial pneumonia, complicated intra-abdominal infections and complicated urinary tract infections.

Methods of Making Pharmaceutical Compositions

[0073] In one aspect, provided herein is a method of preparing a composition comprising ceftolozane and sodium chloride, comprising combining sodium chloride with ceftolozane, wherein 125-1000 mg sodium chloride per 1000 mg of ceftolozane is combined, followed by lyophilization of the sodium chloride ceftolozane mixture.

[0074] In another aspect, provided herein is a method of preparing a composition comprising sodium chloride, tazobactam, and ceftolozane, comprising combining sodium chloride, tazobactam, and ceftolozane, wherein 125-1000 mg sodium chloride per 1000 mg of ceftolozane is combined, followed by lyophilization of the mixture of sodium chloride, tazobactam, and ceftolozane.

[0075] In one embodiment of these methods, 125-500 mg sodium chloride per 1000 mg of ceftolozane is combined.

[0076] In another embodiment of these methods, the method further comprises lyophilizing ceftolozane in the absence of tazobactam. In yet another embodiment of the methods described above, the method can further comprise lyophilizing tazobactam in the absence of ceftolozane.

[0077] In a further embodiment of these methods, the method can comprise the steps of: (1) adding 125 to 1000 mg sodium chloride per 1000 mg of ceftolozane followed by lyophilizing ceftolozane; (2) lyophilizing tazobactam; and (3) combining the separately lyophilized ceftolozane and tazobactam to obtain said pharmaceutical composition. In yet another embodiment, the method comprises adding 125 to 500 mg sodium chloride per 1000 mg of ceftolozane.

[0078] In another aspect, provided herein is a method of making a pharmaceutical composition, comprising combining tazobactam and ceftolozane. In one embodiment, the method further comprises lyophilizing ceftolozane in the absence of tazobactam. In another embodiment, the method further comprises lyophilizing tazobactam in the absence of ceftolozane.

[0079] In a further embodiment, the method comprises the steps of: (1) lyophilizing ceftolozane; (2) lyophilizing tazobactam; and (3) combining the separately lyophilized ceftolozane and tazobactam to obtain said pharmaceutical composition. In one embodiment, the method further comprises packaging the blended powder into Sterbags®.

[0080] In one embodiment of the method, and above embodiments, the molar ratio of tazobactam to ceftolozane in the mixture is in the range of 1:3 to 3:1. In one embodiment, the molar ratio of tazobactam to ceftolozane in the mixture is in the range of 1.2 to 2.1. In one embodiment, the molar ratio of tazobactam to ceftolozane in the mixture is in the range of 1.0 to 0.9:1. In a particular embodiment, the ratio of tazobactam to ceftolozane in the mixture is 0.9:1. In another particular embodiment, the ratio of tazobactam to ceftolozane in the mixture is about 1:2.

Methods of Treatment

[0081] Tazobactam inhibits or decreases the activity of beta-lactamases (e.g., bacterial beta-lactamases), and can be combined with beta-lactam compounds (e.g., antibiotics), thereby broadening the spectrum of the beta-lactam com-
compound and increasing the beta-lactam compound's efficacy against organisms that produce beta-lactamase. A compound or a composition possesses efficacy against an organism if it kills or weakens the organism, or inhibits or prevents reproduction of the organism.

[0082] In one aspect, provided herein is a method for the treatment of bacterial infections in a mammal, comprising administering to said mammal a therapeutically effective amount of a pharmaceutical composition prepared according to the methods described herein. In another aspect, provided herein is a method for the treatment of bacterial infections in a mammal, comprising administering to said mammal a therapeutically effective amount of tazobactam and ceftolozane. In certain embodiments of the above methods, the bacterial infection is caused by an extended-spectrum beta-lactamase-producing organism. In certain embodiments, the bacterial infection is caused by an antibiotic-resistant organism.

[0083] In another aspect, provided herein is a method for the treatment of bacterial infections in a mammal, comprising administering to said mammal a therapeutically effective amount of a pharmaceutical composition comprising ceftolozane. In yet another aspect, the invention is a method for the treatment of bacterial infections in a mammal, comprising administering to said mammal a therapeutically effective amount of a pharmaceutical composition comprising both tazobactam and ceftolozane. In certain embodiments of the above methods, the bacterial infection is caused by an extended-spectrum beta-lactamase-producing organism. In certain embodiments, the bacterial infection is caused by an antibiotic-resistant organism.

[0084] In certain embodiments of both aspects, the pharmaceutical composition further comprises 125 to 1000 mg sodium chloride per 1000 mg of ceftolozane. In some other embodiments, the pharmaceutical composition comprises 125 to 500 mg sodium chloride per 1000 mg of ceftolozane.

[0085] In another aspect, provided herein is a method for the treatment of bacterial infections in a mammal, comprising administering to said mammal a therapeutically effective amount of a pharmaceutical composition comprising tazobactam, ceftolozane, and less than 0.1% by weight of the following compound:

![Chemical Structure](image1)

[0086] In one embodiment of the treatment method, the pharmaceutical composition comprises tazobactam, ceftolozane, and less than 0.05% by weight of the following compound:

![Chemical Structure](image2)
Non-limiting examples of bacterial infections that can be treated by the methods of the invention include infections caused by: aerobic and facultative gram-positive microorganisms (e.g., Staphylococcus aureus, Enterococcus faecalis, Staphylococcus epidermidis, Streptococcus agalactiae, Streptococcus pneumonia, Streptococcus pyogenes, Viridans group streptococci), aerobic and facultative gram-negative microorganisms (e.g., Acinetobacter baumannii, Escherichia coli, Haemophilus influenza, Klebsiella pneumonia, Pseudomonas aeruginosa, Citrobacter koseri, Moraxella catarrhalis, Morganella morganii, Neisseria gonorrhoeae, Proteus mirabilis, Proteus vulgaris, Serratia marcescens, Providencia stuartii, Providencia rettgeri, Salmonella enterica), gram-positive anaerobes (Clostridium perfringens), and gram-negative anaerobes (e.g., Bacteroides fragilis group (e.g., B. fragilis, B. ovatus, B. thetaiotaomicron, and B. vulgatus), Bacteroides distasonis, Prevotella melaninogenicosa).

In certain embodiments of the methods described herein, the bacterial infection resulting from beta-lactamase-producing organisms are treated or controlled. Non-limiting examples of beta-lactamase-producing organisms include:

1. ESBL (extended-spectrum beta-lactamase)-producing organisms selected from the group consisting of Enterobacteriaceae spp. (Escherichia coli, Klebsiella spp., including K. pneumoniae and K. oxytoca), Proteus mirabilis, Proteus vulgaris, Enterobacter spp., Serratia spp., Citrobacter spp., Pseudomonas spp., Acinetobacter spp.) and Bacteroides spp.;

2. CSBL (conventional-spectrum beta-lactamase)-producing organisms, known to those of skill in the art; and

3. Inducible-AmpC-type beta-lactamases, such as Citrobacter spp., Serratia spp., Morganella morganii, Proteus vulgaris, and Enterobacter cloacae.

In certain embodiments of the methods described herein, bacterial infection is associated with one or more of the following conditions:

Community-acquired pneumonia (moderate severity only) caused by piperacillin-resistant, beta-lactamase producing strains of Haemophilus influenza;

Nosocomial pneumonia (moderate to severe) caused by piperacillin-resistant, beta-lactamase producing strains of Staphylococcus aureus and by Acinetobacter baumannii, Haemophilus influenzae, Klebsiella pneumoniae, and Pseudomonas aeruginosa.

Complicated intra-abdominal infections; Complicated urinary tract infections (cUTIs); Acute Pyelonephritis; Systemic Inflammatory Response Syndrome (SIRS).

Also provided herein is the use of tazobactam, and hydrates and solvates thereof, in combination with cefolozane, for the preparation of a medicament for the treatment of bacterial infection. The bacterial infection can result from either gram-negative or gram-positive organisms.

As used herein, “treating”, “treat” or “treatment” describes the management and care of a patient for the purpose of combating a disease, condition, or disorder and includes the administration of a pharmaceutical composition of the present invention to alleviate the symptoms or complications of a disease, condition or disorder, or to eliminate the disease, condition or disorder. The term “treat” can also include treatment of a cell in vitro or an animal model.

By a “therapeutically effective amount” of a compound of the invention is meant a sufficient amount of the compound to treat the disorder (e.g., bacterial infection). The specific therapeutically effective amount that is required for the treatment of any particular patient or organism (e.g., a mammal) will depend upon a variety of factors including the disorder being treated and the severity of the disorder; the activity of the specific compound or composition employed; the specific composition employed; the age, body weight, general health, sex and diet of the patient; the time of administration, route of administration, and rate of excretion of the specific compound employed; the duration of the treatment; drugs used in combination or coincidental with the specific compound employed; and like factors well known in the medical arts (see, for example, Goodman and Gilman’s, “The Pharmacological Basis of Therapeutics”, Tenth Edition, A. Gilman, J. Hardman and L. Limbird, eds., McGraw-Hill Press, 155-173, 2001, which is incorporated herein by reference in its entirety). The therapeutically effective amount for a given situation can be readily determined by routine experimentation and is within the skill and judgment of the ordinary clinician.

EXAMPLES

Example 1

Manufacturing Procedure of Mono Product for Injection

1. Preparation of the Compound Solution of CXA-101 Lyophilized Product

1. Weigh 30 kg of water for injection into the compounding vessel;

2. Add 100 g of citric acid, anhydrous and 150 g of sodium bicarbonate into the compounding vessel and dissolve them with mixing;

3. Weigh 5,000 g potency of CXA-101 drug substance and suspend it with mixing. (Note any generation of carbon dioxide.)

4. Slowly add 1,100 g of sodium bicarbonate and dissolve CXA-101 with mixing. (Again, note any generation of carbon dioxide.)

5. Add 1,146 g of sodium chloride and 10,000 g of maltose, dissolve with mixing.

6. Purge dissolved carbon dioxide in the solution with nitrogen until the pH of the solution does not change.

7. Adjust the pH of the solution to 6.0±0.1 with 5%-sodium bicarbonate solution.

8. Adjust the total weight to 56,850 g (D_{20}=-1.137) with water for injection.

9. Confirm the pH of the compound solution within the range 6.0±0.1.

1.2. Prefiltration and Sterile-Filtration

10. Filter the compounded solution with a sterile filter-set which consists of a 0.2 um polyvinylidene fluoride membrane filter (Durapore®, Millipore) and a 0.1 um polyvinylidene fluoride membrane filter (Durapore®, Millipore) connected in tandem. Confirm the integrity of each filter before and after the filtration. Take approximately 100 mL of the filterate in order to check bioburden.

11. Filter the prefiltered compounded solution through a sterile filter-set which consists of a 0.2 um polyvinylidene fluoride membrane filter and a 0.1 um polyvinylidene fluoride membrane filter connected in tandem, and
introduce the final filtrate into an aseptic room. Confirm the integrity of each filter before and after the filtration.

1.3. Processing of Vial, Stopper and Flip-Off Cap

   12) Wash a sufficient quantity of 28 mL vials with water for injection and sterilize the washed vials by a dry-heat sterilizer. Then transfer the sterilized vials into a Grade A area located in an aseptic room.

   13) Wash a sufficient quantity of stoppers with water for injection. Sterilize and dry the washed stoppers by steam sterilizer. Then transfer the sterilized stoppers into a Grade A area located in an aseptic room.

   14) Sterilize a sufficient quantity of flip-off caps by steam sterilizer. Then transfer the sterilized flip-off caps into a Grade A or B area located in an aseptic room.

1.4. Filling and Partially Stopping

   15) Adjust the fill weight of the filtered compounded solution to 11.37 g (corresponds to 10 mL of the compounded solution), then start filling operation. Check the filled weight in sufficient frequency and confirm it is in target range (11.37 g±1%, 11.26 to 11.43 g). When deviation from the control range (11.37 g±2%, 11.14 to 11.59 g) is occurred, re-adjust the filling weight.

   16) Immediately after a vial is filled, partially stopper the vial with a sterilized stopper. Load the filled and partially stoppered vials onto the shelves of a lyophilizer aseptically.

1.5. Lyophilization to Crimping, Visual Inspection, Labeling and Packaging

   17) After all filled and partially stoppered vials are loaded into a lyophilizer, start the lyophilization program shown in FIG. 1. Freeze the loaded vials at −40°C, and keep until all vials freeze. Forward the program to primary drying step (shelf temperature: −20°C, chamber pressure: 100 to 150 mTorr). Primary drying time should be determined by monitoring the product temperature. Forward the program to secondary drying step (shelf temperature: 30°C, chamber pressure: not more than 10 mTorr) after completion of the primary drying step. After all vials are dried completely, return the chamber pressure to atmospheric pressure with sterilized nitrogen. Then stopper vials completely.

   18) Unload the lyophilized vials from the chamber and crimp with sterilized flip-off caps.

   19) Subject all crimped vials to visual inspection and label and package all passed vials.

Example 2

   Manufacturing Procedure of Bulk (Tray) Lyophilized Cefazolozane

   There are four main steps in the manufacture of CXA-101 bulk drug product: dissolution, sterile filtration, bulk lyophilization, and packaging into Sterbags®. These four main steps are composed of a total of 20 minor steps. The flowchart of the CXA-101 bulk drug product manufacturing process is presented in FIG. 3, and described below.

   I. Dissolution

   1. The prescribed amount of WFI is charged into the dissolution reactor.

   2. A prescribed amount of citric acid is added.

   3. The solution is cooled at 5°C to 10°C.

   4. A prescribed amount of CXA-101 drug substance is added to the solution.

   5. A prescribed amount of L-arginine is slowly added to the solution.

   6. A check for complete dissolution is performed. Solution pH is verified to be in the target range of 6.5 to 7.0.

   7. A prescribed amount of sodium chloride is added to the solution.

   8. A check for complete dissolution is performed. Solution pH is verified to be in the target range of 6.0 to 7.0. If the pH is out of this range adjust with either L-Arginine or citric acid.

   9. WFI is added to bring the net weight to 124.4 kg and the solution is mixed well.

   10. Samples are withdrawn for testing of final pH.

   II. Sterile Filtration

   11. The solution is passed through the filter (pore size 0.45 μm) followed by double filters (pore size 0.22 μm) onto a shelf on the Criofarma lyophilizer.

   12. The line is washed with WFI.

   13. The washing solution is passed from Step 12 through sterile filtration.

   III. Bulk Lyophilization

   14. The washing solution is loaded onto a separate shelf in the lyophilizer (and later discarded).

   15. The solution is lyophilized until dry.

   16. The product shelf is cooled to 20°C ±5°C.

   IV. Packaging into Sterbags®

   17. The lyophilized bulk drug product powder is milled.

   18. The milled powder is sieved.

   19. The sieved powder is blended for 30 minutes.

   20. The powder is then discharged into Sterbags®.

Example 3

   Manufacturing of Combination Product (Tazobactam and CXA-101) by Blending

A. Sterile Dry Blending of Bulk Lyophilized Cefazolozane and Bulk Lyophilized Tazobactam

   A low energy drum blender that agitates the material by tumbling and also moving the bed up and down is used. A representative process of blending is described as follows. For CXA-101/tazobactam for injection, the blender was charged with 23.4 kg of CXA-101 bulk product, and 5.4 kg of tazobactam bulk product. Both the CXA-101 and tazobactam were individually lyophilized beforehand. The material was blended for 180 minutes. In-process tests of content assay for both CXA-101 and tazobactam were performed to assess the homogeneity using the samples of blend materials taken from three places. The RSD for each of CXA-101 and tazobactam content assay was no greater than 2% and the RSD for the ratio of CXA-101/tazobactam was no greater than 2% (See Table 1).
TABLE 1

In-Process Testing of Blending Samples of a CXA-201 Composition at Three Places

<table>
<thead>
<tr>
<th>Acceptance Limits (expected value)</th>
<th>Sampling</th>
<th>60</th>
<th>120</th>
<th>180</th>
</tr>
</thead>
<tbody>
<tr>
<td>Test</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Content: 30.4%-37.2%</td>
<td>1</td>
<td>34.24</td>
<td>34.07</td>
<td>34.42</td>
</tr>
<tr>
<td>Ceflozole</td>
<td>2</td>
<td>34.62</td>
<td>34.21</td>
<td>34.66</td>
</tr>
<tr>
<td>Tazobactam: 15.2%-18.0%</td>
<td>1</td>
<td>17.96</td>
<td>18.20</td>
<td>17.12</td>
</tr>
<tr>
<td>Cap apez</td>
<td>3</td>
<td>16.90</td>
<td>18.26</td>
<td>16.51</td>
</tr>
<tr>
<td>Mean</td>
<td>3</td>
<td>34.52</td>
<td>34.30</td>
<td>34.64</td>
</tr>
<tr>
<td>RSD %</td>
<td></td>
<td>0.72</td>
<td>0.80</td>
<td>0.63</td>
</tr>
<tr>
<td>Ratio of</td>
<td>2.00%</td>
<td>1</td>
<td>1.91</td>
<td>1.87</td>
</tr>
<tr>
<td>Content (w/w)</td>
<td>2</td>
<td>2.05</td>
<td>1.87</td>
<td>2.10</td>
</tr>
<tr>
<td>Ceflozole/tazobactam</td>
<td>3</td>
<td>2.01</td>
<td>2.04</td>
<td>2.05</td>
</tr>
<tr>
<td>Mean</td>
<td></td>
<td>1.99</td>
<td>1.93</td>
<td>2.05</td>
</tr>
<tr>
<td>RSD %</td>
<td></td>
<td>2.69</td>
<td>5.12</td>
<td>2.2</td>
</tr>
</tbody>
</table>

RSD = relative standard deviation

1) Theoretical value: 33.96% Acceptance limits are 90%-110% of the theoretical value.
2) Theoretical value: 16.99% Acceptance limits are 90%-110% of the theoretical value.

Example 4

Analytical HPLC Method

A. Operative Conditions

Column: Develosil ODS-UG-5, 5 μm, 250 x 4.6 mm (Nomura Chemical, Japan)
Mobile phase: Sodium Perchlorate Buffer Solution (pH 2.5)/CH3CN 90:10 (v/v)
Flow rate: 1.0 mL/min
Injection volume: 10 μL
Oven temperature: 45°C
Run Time: 85 minutes

Gradient Profile:

<table>
<thead>
<tr>
<th>Time (min)</th>
<th>A %</th>
<th>B %</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>75</td>
<td>25</td>
</tr>
<tr>
<td>30</td>
<td>70</td>
<td>30</td>
</tr>
<tr>
<td>60</td>
<td>0</td>
<td>100</td>
</tr>
<tr>
<td>85</td>
<td>0</td>
<td>100</td>
</tr>
<tr>
<td>85.1</td>
<td>75</td>
<td>25</td>
</tr>
<tr>
<td>110</td>
<td>75</td>
<td>25</td>
</tr>
</tbody>
</table>

B. Mobile Phase Preparation

Sodium perchlorate buffer solution was made by dissolving 14.05 g of sodium perchlorate monohydrate in 1000.0 mL of water followed by adjusting pH to 2.5 with diluted perchloric acid (1 in 20).

Mobile phase was then made by mixing sodium perchlorate buffer solution (pH 2.5) and acetonitrile in the ratio 90:10 (v/v).

Sodium acetate buffer solution pH 5.5 (dilute) was made by dissolving 1.36 g of sodium acetate trihydrate in 1000.0 mL of water followed by adjusting to pH 5.5 with diluted acetic acid (1 in 10).

C. Sample Preparation

Sample solution: dissolve 20.0 mg, exactly weighed, of a sample, in 20.0 mL of water (prepare just before injection into HPLC system).

System suitability solution (1%) take 1.0 mL of the sample solution (use first sample if more are present) and transfer into a 100.0 mL volumetric flask, dilute with water to volume and mix.

D. HPLC Analysis Procedure

1. Inject blank (water)

2. Inject system suitability solution and check for tailing factor and theoretical plate number for CXA-101 peak:

Theoretical plates number must not be greater than 1.5

3. Inject sample solution
4. Inject system suitability solution and check for tailing factor and theoretical plate number for CXA-101 peak.

The tailing factor must not be greater than 1.5

Theoretical plates number must not be less than 10000

5. Identify the peaks of Related Substances in the Sample chromatogram based on the reference chromatogram reported in FIG. 5 or, alternatively, on the basis of the following RRT values:

<table>
<thead>
<tr>
<th>Impurity</th>
<th>RRT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peak 1 (P1)</td>
<td>~0.1</td>
</tr>
<tr>
<td>Peak 2 (P2)</td>
<td>~0.2</td>
</tr>
<tr>
<td>Peak 3 (P3)</td>
<td>~0.4</td>
</tr>
<tr>
<td>Peak 4 (P4)</td>
<td>~0.6</td>
</tr>
<tr>
<td>Peak 5 (P5)</td>
<td>~0.9</td>
</tr>
<tr>
<td>CXA-101</td>
<td>1.0</td>
</tr>
<tr>
<td>Peak 6 (P6)</td>
<td>~1.1</td>
</tr>
<tr>
<td>Peak 7 (P7)</td>
<td>~1.3</td>
</tr>
<tr>
<td>Peak 8 (P8)</td>
<td>~1.4</td>
</tr>
<tr>
<td>Peak 9 (P9)</td>
<td>~1.7</td>
</tr>
<tr>
<td>Peak s 10, 11 (P10, 11)</td>
<td>~2.3</td>
</tr>
</tbody>
</table>

E. Calculations

I. Report for Each Related Substance its Amount as Expressed by Area Percent.

\[ C_i = \frac{A_i \times 100}{A_i + \Sigma A_i} \]

wherein:

\[ C_i \] = Amount of related substance \( i \) in the Sample, area %

\[ A_i \] = Peak area of related substance \( i \) in the Sample chromatogram

\[ A_i + \Sigma A_i \] = Area of CXA-101 peak in the Sample chromatogram

Consider as any Unspecified Impurity, each peak in the chromatogram except CXA-101, peaks from 1 to 11 and every peak present in the blank chromatogram and report the largest.

II. Report the Total Impurities Content as Expressed by the Following Formula:

\[ C_T = \frac{A_T \times 100}{A_T + \Sigma A_i} \]

wherein:

\[ C_T \] = total impurities content in the Sample, area %

\[ A_T \] = area of CXA-101 peak in the sample chromatogram

\[ \Sigma A_i \] = total peak areas of impurities in the sample chromatogram

Example 5
Stabilizing Effect of Sodium Chloride

A. Reduction of the Impurity at RT=63 minutes

A stability study was carried out at 25°C, and samples were analyzed by HPLC. High, mid, and low salt formulations contained 480, 125, and 62.5 mg NaCl per 1000 mg of ceftolozane, respectively. Compositions of blend Drug Product are listed in Table 2. Test results are summarized in Table 3.

<table>
<thead>
<tr>
<th>TABLE 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Comparison of the CXA-101 Compositions</td>
</tr>
<tr>
<td>Lot</td>
</tr>
<tr>
<td>-----</td>
</tr>
<tr>
<td>C1</td>
</tr>
<tr>
<td>C2</td>
</tr>
<tr>
<td>C3</td>
</tr>
<tr>
<td>C4</td>
</tr>
<tr>
<td>C5</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>TABLE 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>RT 63° Peak Area at t = 3 months, 25°C/60% RH storage</td>
</tr>
<tr>
<td>Sample</td>
</tr>
<tr>
<td>--------</td>
</tr>
<tr>
<td>C1</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>C2</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>C3</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>C4</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>C5</td>
</tr>
<tr>
<td></td>
</tr>
</tbody>
</table>

Conclusion:

at the three month time point, the reduced salt formulations were observed to be as stable as the full salt formulation; and trends indicate that reduction in salt causes at least 1.5-fold greater impurity at RT=63 minutes (HPLC). CXA-101 Peak Trends with NaCl

A stability study was carried out at 30°C and 60°C, and samples were analyzed by HPLC. Sodium chloride content in test samples is described in Table 4. Stability data are summarized in Tables 5-8. The data are also plotted in FIGS. 6-9 to show trends of total purity, peak 1, RRT 0.43+ peak 3, and peak 7 with respect to NaCl.

<table>
<thead>
<tr>
<th>TABLE 4</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sodium Chloride Content in the CXA-101 Compositions</td>
</tr>
<tr>
<td>Samples</td>
</tr>
<tr>
<td>---------</td>
</tr>
<tr>
<td>A1</td>
</tr>
<tr>
<td>A2</td>
</tr>
<tr>
<td>A3</td>
</tr>
<tr>
<td>A4</td>
</tr>
<tr>
<td>A5</td>
</tr>
</tbody>
</table>
TABLE 5
The Purity of Ceftolozane in CXA-101 Compositions with Varying Amounts of Sodium Chloride

<table>
<thead>
<tr>
<th>Day A1 A2 A3 A4 A5</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>70/60° C.</td>
<td>0 96.6 98.0 97.9 97.8 97.7</td>
</tr>
<tr>
<td>70/30° C.</td>
<td>0 98.1 97.8 97.8 97.7</td>
</tr>
<tr>
<td>1 day/60° C.</td>
<td>1 95.9 96.9 96.5 95.7 95.5</td>
</tr>
<tr>
<td>1 day/30° C.</td>
<td>1 98.2 97.7 97.7 97.6</td>
</tr>
<tr>
<td>3 days/60° C.</td>
<td>3 94.9 95.7 94.8 93.9 93.6</td>
</tr>
<tr>
<td>(Δt0)</td>
<td>(1.7) (2.3) (3.1) (3.9) (4.1)</td>
</tr>
<tr>
<td>3 day/30° C.</td>
<td>3 98.0 97.5 97.5 97.3</td>
</tr>
<tr>
<td>7 days/60° C.</td>
<td>7 93.6 94.0 94.2 92.3 91.9</td>
</tr>
<tr>
<td>7 day/30° C.</td>
<td>7 97.8 97.2 97.1 97.0</td>
</tr>
<tr>
<td>Total ∆/60° C.</td>
<td>3.07 4.06 3.7 5.48 5.83</td>
</tr>
<tr>
<td>Total ∆/30° C.</td>
<td>0.3 0.6 0.7 0.7 0.7</td>
</tr>
</tbody>
</table>

TABLE 6
The HPLC Peak Area of Impurity Peak 1 in CXA-101 Compositions with Varying Amounts of Sodium Chloride

<table>
<thead>
<tr>
<th>Day A1 A2 A3 A4 A5</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>70/60° C.</td>
<td>0 0.95 0.31 0.3 0.36 0.39</td>
</tr>
<tr>
<td>70/30° C.</td>
<td>0 0.47 0.36 0.36 0.39</td>
</tr>
<tr>
<td>1 day/60° C.</td>
<td>1 1.36 0.86 0.94 1.36 1.39</td>
</tr>
<tr>
<td>1 day/30° C.</td>
<td>1 0.48 0.40 0.42 0.48</td>
</tr>
<tr>
<td>3 days/60° C.</td>
<td>3 1.71 1.31 1.73 2.06 2.1</td>
</tr>
<tr>
<td>3 day/30° C.</td>
<td>3 0.53 0.50 0.52 0.58</td>
</tr>
<tr>
<td>7 days/60° C.</td>
<td>7 2.26 2.14 2.07 2.86 2.93</td>
</tr>
<tr>
<td>7 day/30° C.</td>
<td>7 0.62 0.63 0.66 0.72</td>
</tr>
<tr>
<td>INCREASE %/ 60° C.</td>
<td>1.31 1.83 1.77 2.5 2.54</td>
</tr>
<tr>
<td>INCREASE %/ 30° C.</td>
<td>0.15 0.27 0.3 0.33</td>
</tr>
</tbody>
</table>

TABLE 7
The Total HPLC Peak Area of the Impurity with a RRT of 0.43 and Impurity peak 3 in CXA-101 Compositions with Varying Amounts of Sodium Chloride

<table>
<thead>
<tr>
<th>Day A1 A2 A3 A4 A5</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>70/60° C.</td>
<td>0 0.28 0.10 0.09 0.10 0.11</td>
</tr>
<tr>
<td>70/30° C.</td>
<td>0 0.15 0.10 0.10 0.11</td>
</tr>
<tr>
<td>1 day/60° C.</td>
<td>1 0.37 0.13 0.16 0.35 0.36</td>
</tr>
<tr>
<td>1 day/30° C.</td>
<td>1 0.13 0.09 0.09 0.10</td>
</tr>
<tr>
<td>3 days/60° C.</td>
<td>3 0.68 0.21 0.31 0.71 0.71</td>
</tr>
<tr>
<td>3 day/30° C.</td>
<td>3 0.17 0.13 0.13 0.14</td>
</tr>
<tr>
<td>7 days/60° C.</td>
<td>7 1.04 0.36 0.31 0.81 0.81</td>
</tr>
<tr>
<td>7 day/30° C.</td>
<td>7 0.19 0.16 0.16 0.17</td>
</tr>
<tr>
<td>INCREASE %/ 60° C.</td>
<td>0.76 0.26 0.21 0.71 0.7</td>
</tr>
<tr>
<td>INCREASE %/ 30° C.</td>
<td>0.04 0.06 0.06 0.06</td>
</tr>
</tbody>
</table>

TABLE 8
The HPLC Peak Area of Impurity Peak 7 in CXA-101 Compositions with Varying Amounts of Sodium Chloride

<table>
<thead>
<tr>
<th>Day A1 A2 A3 A4 A5</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>70/60° C.</td>
<td>0 1.31 0.95 0.96 1.01 1.02</td>
</tr>
<tr>
<td>70/30° C.</td>
<td>0 0.69 1.00 1.01 1.02</td>
</tr>
<tr>
<td>1 day/60° C.</td>
<td>1 1.37 1.10 1.10 1.23 1.29</td>
</tr>
<tr>
<td>1 day/30° C.</td>
<td>1 0.68 0.99 1.01 1.02</td>
</tr>
<tr>
<td>3 days/60° C.</td>
<td>3 1.43 1.19 1.27 1.41 1.46</td>
</tr>
</tbody>
</table>

TABLE 9
The Sodium Chloride Content in the CXA-101 Compositions

<table>
<thead>
<tr>
<th>Samples</th>
<th>NaCl content</th>
</tr>
</thead>
<tbody>
<tr>
<td>B1</td>
<td>481.0 mg sodium chloride per 1000 mg of ceftolozane</td>
</tr>
<tr>
<td>B2</td>
<td>125.0 mg sodium chloride per 1000 mg of ceftolozane</td>
</tr>
<tr>
<td>B3</td>
<td>75.0 mg sodium chloride per 1000 mg of ceftolozane</td>
</tr>
<tr>
<td>B4</td>
<td>50.0 mg sodium chloride per 1000 mg of ceftolozane</td>
</tr>
</tbody>
</table>

TABLE 10
The Purity of Ceftolozane in CXA-201 Compositions with Varying Amounts of Sodium Chloride

<table>
<thead>
<tr>
<th>Day A1 A2 A3 A4 A5</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>70/60° C.</td>
<td>0 98.1 97.8 97.8 97.7</td>
</tr>
<tr>
<td>1 day/60° C.</td>
<td>1 97.2 96.3 96.2 96.0</td>
</tr>
<tr>
<td>1 day/30° C.</td>
<td>1 98.2 97.7 97.6 97.6</td>
</tr>
<tr>
<td>3 days/60° C.</td>
<td>3 95.4 94.9 94.7 94.6</td>
</tr>
<tr>
<td>(Δt0)</td>
<td>(2.7) (2.9) (3.1) (3.1)</td>
</tr>
<tr>
<td>3 day/30° C.</td>
<td>3 98.0 97.5 97.4 97.3</td>
</tr>
<tr>
<td>7 days/60° C.</td>
<td>7 92.7 93.8 93.6 93.4</td>
</tr>
<tr>
<td>7 day/30° C.</td>
<td>7 97.8 97.2 97.0 96.9</td>
</tr>
<tr>
<td>Total ∆/60° C.</td>
<td>5.3 4.0 4.2 4.3</td>
</tr>
<tr>
<td>Total ∆/30° C.</td>
<td>0.3 0.6 0.8 0.8</td>
</tr>
</tbody>
</table>

TABLE 11
The HPLC Peak Area of Impurity Peak 1 of Ceftolozane in CXA-201 Compositions with Varying Amounts of Sodium Chloride

<table>
<thead>
<tr>
<th>Day A1 A2 A3 A4 A5</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>t0</td>
<td>0 0.47 0.38 0.38 0.41</td>
</tr>
<tr>
<td>1 day/60° C.</td>
<td>1 1 1.08 1.09 1.14</td>
</tr>
<tr>
<td>1 day/30° C.</td>
<td>1 0.48 0.44 0.45 0.49</td>
</tr>
<tr>
<td>3 days/60° C.</td>
<td>3 1.85 1.64 1.66 1.71</td>
</tr>
<tr>
<td>3 day/30° C.</td>
<td>3 0.53 0.53 0.56 0.61</td>
</tr>
<tr>
<td>7 days/60° C.</td>
<td>7 3.3 2.28 2.25 2.29</td>
</tr>
<tr>
<td>7 day/30° C.</td>
<td>7 0.62 0.67 0.71 0.77</td>
</tr>
</tbody>
</table>

[0172] Conclusion:
C. CXA-201 Peak Trends with NaCl
[0174] A stability study was carried out at 30° C and 60° C, and samples were analyzed by HPLC. Sodium chloride content is described in Table 9. Stability data at 60° C are summarized in Tables 10-13. The data are also plotted in FIGS. 10-13 to show trends of total purity, peak 1, RRT 0.43+peak 3, and peak 7 with respect to NaCl.
Conclusion:

The stability data shows that high sodium chloride content enhances stability of Combination product CXA-201.

Example 6

Manufacturing of Combination Product (Tazobactam and CXA-101) by Co-Lyophilization

0176 The co-lyophilization process is illustrated in FIG. 14. The components of a CXA-201 composition prepared by co-lyophilization are described in Table 14 below.

Example 7

Assessment of Co-Lyophilized Combo Drug Product

A. Preparation of Co-Lyophilized Combo Drug Product

0177 The Co-Iyo Combo Drug Product was prepared, as described above in Example 6. The formulation composition of the Combo drug product is shown in Table 15.

B. Stress Test

0178 This sample was put into stability study. The following Tables 16 and 17 are representative examples that summarize the results at 25°C/RH=60% and 40°C/RH=75% after one month (T1) and three months (T2). Samples were analyzed using the HPLC method as described in Example 4.
TABLE 16

Stability Data of Co-Lyophilized CXA-201 Composition at 25° C/RH = 60%

<table>
<thead>
<tr>
<th>Test items</th>
<th>Spec. D.P.</th>
<th>T0</th>
<th>T1 25° C.</th>
<th>T2 25° C.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Related Substances</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Peak 1</td>
<td>≤1.50%</td>
<td>0.31%</td>
<td>0.54%</td>
<td>0.71%</td>
</tr>
<tr>
<td>Peak 2</td>
<td>≤0.40%</td>
<td>0.07%</td>
<td>0.07%</td>
<td>0.09%</td>
</tr>
<tr>
<td>Peak 3</td>
<td>≤0.30%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Peak 4</td>
<td>≤0.80%</td>
<td>0.08%</td>
<td>0.08%</td>
<td>0.09%</td>
</tr>
<tr>
<td>Peak 5</td>
<td>≤1.00%</td>
<td>0.27%</td>
<td>0.26%</td>
<td>0.29%</td>
</tr>
<tr>
<td>Peak 6</td>
<td>≤0.15%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Peak 7</td>
<td>≤2.00%</td>
<td>0.64%</td>
<td>0.65%</td>
<td>0.66%</td>
</tr>
<tr>
<td>Peak 8</td>
<td>≤0.15%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Peak 9</td>
<td>≤0.60%</td>
<td>0.05%</td>
<td>0.11%</td>
<td>0.10%</td>
</tr>
<tr>
<td>Peak 10, 11</td>
<td>≤0.15% each</td>
<td>0.04%</td>
<td>0.04%</td>
<td>0.04%</td>
</tr>
<tr>
<td>Peak 12</td>
<td>≤2.00%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Others (RRT 0.43)</td>
<td>≤0.15%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Others (RRT 1.22)</td>
<td>≤0.15%</td>
<td>0.13%</td>
<td>0.30%</td>
<td>0.38%</td>
</tr>
<tr>
<td>Others (RRT 2.18)</td>
<td>≤0.15%</td>
<td>0.03%</td>
<td>&lt;0.03%</td>
<td>0.05%</td>
</tr>
<tr>
<td>Others (RRT 2.77)</td>
<td>≤0.15%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Sing. Unk.</td>
<td>≤0.15%</td>
<td>0.05%</td>
<td>0.07%</td>
<td>0.05%</td>
</tr>
<tr>
<td>Total</td>
<td>≤5.00%</td>
<td>1.67%</td>
<td>2.19%</td>
<td>2.77%</td>
</tr>
<tr>
<td>pH</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

C. Conclusion:

[0179] A new impurity having RRT=1.22 was observed in the co-lyophilized drug product. The impurity was identified as a degradation product, shown in FIG. 15, which was formed by a reaction between cefotaxime and formylacetic acid, which was a degradation product of tazobactam. The stability data at 25° C. and at 40° C. have shown that the impurity increases over time.

Example 8

Assessment of Blend Combination Drug Product

A. Preparation of Blend Combination Drug Product

[0180] The blend drug product was prepared, as described above in Example 3, on lab scale by use of a small blender. The composition of the blend drug product is shown in Table 18.

**TABLE 18**

Components of the Blend Composition

<table>
<thead>
<tr>
<th>Component</th>
<th>Composition</th>
<th>Quantity as active components</th>
</tr>
</thead>
<tbody>
<tr>
<td>CXA-201</td>
<td>Cefotaxime</td>
<td>10.8 g</td>
</tr>
<tr>
<td>Comp. 1</td>
<td>L-Arginine</td>
<td>6.7 g</td>
</tr>
<tr>
<td>101 for Injection</td>
<td>Citric acid</td>
<td>233 mg</td>
</tr>
<tr>
<td>Bulk (25)</td>
<td>Sodium chloride</td>
<td>5.2 g (as Tazo sterile Bulk (6 g) free acid)</td>
</tr>
</tbody>
</table>

B. Stress Test

[0181] This sample was put into stability study. The following Tables 19 and 20 are representative examples that summarize the results at 25° C/RH=60% and 40° C/RH=75% after one month (T1) and three months (T2). Samples were analyzed using the HPLC method as described in Example 4.

**TABLE 19**

Stability Data of Blend CXA-201 Composition at 25° C/RH = 60%

<table>
<thead>
<tr>
<th>Test items</th>
<th>Specifications</th>
<th>T0</th>
<th>T1 25° C.</th>
<th>T2 25° C.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Related Substances</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Peak 1</td>
<td>≤1.50%</td>
<td>0.11%</td>
<td>0.13%</td>
<td>0.14%</td>
</tr>
<tr>
<td>Peak 2</td>
<td>≤0.40%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Peak 3</td>
<td>≤0.15%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Peak 4</td>
<td>≤1.00%</td>
<td>0.13%</td>
<td>0.14%</td>
<td>0.15%</td>
</tr>
<tr>
<td>Peak 5</td>
<td>≤0.15%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Peak 6</td>
<td>≤0.15%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Peak 7</td>
<td>≤0.20%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Peak 8</td>
<td>≤0.15%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Peak 9</td>
<td>≤0.15%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Peak 10, 11</td>
<td>≤0.15%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
<td>&lt;0.03%</td>
</tr>
<tr>
<td>Sing. Unk.</td>
<td>≤0.15%</td>
<td>0.13%</td>
<td>0.13%</td>
<td>0.13%</td>
</tr>
<tr>
<td>Total</td>
<td>≤5.00%</td>
<td>2.49%</td>
<td>3.03%</td>
<td>3.28%</td>
</tr>
<tr>
<td>pH</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
C. Conclusion

The data at both 25°C and at 40°C have shown that the blending process completely inhibits formation of the impurity RRT=1.22.

1. A composition comprising tazobactam combined with ceftolozane that is lyophilized in the absence of tazobactam.

2. The composition of claim 1, wherein the lyophilized ceftolozane is ceftolozane sulfate.

3. The composition of claim 1, wherein the lyophilized ceftolozane is a ceftolozane compound of formula (I) prior to lyophilization:

\[
\text{Ceftolozane (I)}
\]

4. The composition of claim 3, wherein the lyophilized ceftolozane is obtained by a process comprising:
   a. forming a solution comprising the compound of formula (I) without tazobactam;
   b. adjusting the pH of the solution to a pH of about 6.0; and
   c. lyophilizing the solution to obtain the lyophilized ceftolozane.

5. The composition of claim 1, wherein the composition has a pH of 6.0 upon reconstitution with saline.

6. The composition of claim 1, comprising about 30.4%-37.2% by weight ceftolozane active.

7. The composition of claim 1, comprising about 15.2%-18.6% by weight tazobactam active.

8. The composition of claim 1, comprising ceftolozane and tazobactam (as free acid) in a 2:1 weight ratio.

9. The composition of claim 1, comprising (6R,7R)-3-[5-Amino-4-[3-(2-aminoethyl)ureido]-1-methyl-1H-pyrazol-2-ium-2-ylmethyl]-7-[2-(5-aminoo-1,2,4-thiadiazol-3-yl)-2-[(Z)-1-carboxyl-1-methylethoxyiminoo]acetamido]-3-cephem-4-carboxylic acid) and tazobactam (as free acid) in a about 2:1 weight ratio.

10. The composition of claim 1, comprising (6R,7R)-3-[5-amino-4-[(2-aminoethyl)carbamoylamino]-1-methyl-1H-pyrazol-2-ium-2-ylmethyl]-7-((2Z)-2-(5-amino-1,2,4-thiadiazol-3-yl)-2-[(1-carboxyl-1-methylethoxyiminoo]acetyl)amino)-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylate and tazobactam (as free acid) in a about 2:1 weight ratio.

11. A ceftolozane/tazobactam composition comprising ceftolozane combined with tazobactam, wherein the ceftolozane and the tazobactam are individually freeze-dried prior to combination.

12. The ceftolozane/tazobactam composition of claim 11, wherein the ceftolozane and the tazobactam are blended to form a ceftolozane/tazobactam composition comprising about 30.4%-37.2% by weight ceftolozane active and about 15.2%-18.6% by weight tazobactam (as free acid).

13. The ceftolozane/tazobactam composition of claim 11, wherein the ceftolozane is ceftolozane sulfate.

14. The ceftolozane/tazobactam composition of claim 11, wherein the ceftolozane is obtained by a process comprising freeze-drying an aqueous solution comprising the ceftolozane in the absence of tazobactam at a pH of 6.0-7.0.

15. The ceftolozane/tazobactam composition of claim 11, wherein the composition has a pH of about 6.0 upon reconstitution in saline.

16. The ceftolozane/tazobactam composition of claim 11 in a unit dosage form comprising a total of 1000 mg ceftolozane and a total of 500 mg tazobactam (as free acid) in a unit dosage form.

17. The ceftolozane/tazobactam composition of claim 11, wherein the lyophilized ceftolozane is a ceftolozane compound of formula (I) prior to lyophilization:
18. A ceftolozane/tazobactam composition comprising ceftolozane combined with tazobactam in a 2:1 weight ratio between the ceftolozane active and the tazobactam active, wherein the ceftolozane is obtained from a solution comprising ceftolozane in the absence of tazobactam.

19. The ceftolozane/tazobactam composition of claim 18, wherein the ceftolozane in the solution is a ceftolozane compound of formula (I):

\[
\text{H}_2\text{N}\text{-}\text{S}-\text{N}\text{-}\text{O}\text{-}\text{H}\text{-}\text{N}_{\text{H}}\text{N}\text{S}-\text{N}\text{-}\text{O}\text{-}\text{CN}_{\text{H}}\text{H}_{\text{N}}\text{-}\text{H}\text{N}\text{-}\text{CO}_{2}\text{H}
\]

and adjusting the pH of the ceftolozane solution to about 6.0 prior to lyophilization.

20. The ceftolozane/tazobactam composition of claim 18, wherein the ceftolozane is obtained by lyophilizing the solution in the absence of tazobactam.

21. The ceftolozane/tazobactam composition of claim 20, wherein the pH of the solution is adjusted to about 6.0 prior to lyophilization.

22. The ceftolozane/tazobactam composition of claim 18, obtained by a process comprising the steps of:
   a. lyophilizing the solution comprising ceftolozane in the absence of tazobactam to obtain a lyophilized ceftolozane composition; and
   b. combining the lyophilized ceftolozane composition and with tazobactam to form the pharmaceutical composition.

23. The pharmaceutical composition of claim 22, wherein the ceftolozane is ceftolozane sulfate.

24. The pharmaceutical composition of claim 22, wherein the tazobactam is tazobactam sodium.

25. The pharmaceutical composition of claim 22, comprising lyophilizing the ceftolozane at a pH of about 6.0.

26. The composition of claim 25, comprising forming the solution with a ceftolozane compound of formula (I):

\[
\text{H}_2\text{N}\text{-}\text{S}-\text{N}\text{-}\text{O}\text{-}\text{H}\text{-}\text{N}_{\text{H}}\text{N}\text{S}-\text{N}\text{-}\text{O}\text{-}\text{CN}_{\text{H}}\text{H}_{\text{N}}\text{-}\text{H}\text{N}\text{-}\text{CO}_{2}\text{H}
\]

27. A pharmaceutical composition obtained by:
   a. adjusting the pH of a solution comprising a compound of formula (I) to a pH of about 5.0-7.0 in the absence of tazobactam;
   b. freeze-drying the solution to obtain a ceftolozane composition; and
   c. combining the ceftolozane composition and with tazobactam to form the pharmaceutical composition.

28. (canceled)

29. The pharmaceutical composition of claim 27, wherein the ceftolozane composition is ceftolozane sulfate.

30. The pharmaceutical composition of claim 29, wherein the lyophilized tazobactam is tazobactam sodium.

31. The pharmaceutical composition of claim 29, wherein the pharmaceutical composition comprises ceftolozane active and tazobactam active in a 2:1 weight ratio.

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