Zosyn® (piperacillin/tazobactam) reformulation: Expanded compatibility and coadministration with lactated Ringer's solutions and selected aminoglycosides

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Abstract: Zosyn®, also known as Tazocin® for injection, contains piperacillin and tazobactam and was approved by the US Food and Drug Administration in 1993 for the treatment of indicated serious infections. In 1995, United States Pharmacopoeia and European Pharmacopoeias reduced the particulate limit for injectables by 40%, based on general safety concerns. Wyeth attempted to control sporadic batch failures (associated with increased particulate formation) by shortening product expiration dating from 36 to 24 months and optimizing the stopper siliconization process. These modifications did not correct the problem completely. Wyeth reformulated Zosyn by incorporating two stabilizing functional excipients, ethylene diamine tetraacetic acid disodium salt (EDTA disodium) and sodium citrate, which solved the particulate formation problem. These two functional excipients also allowed for the first time Y-site coadministration of reformulated Zosyn product with amikacin and gentamicin at specific doses and concentrations, and with certain diluents, and the use of Ca^{++} ion-containing Lactated Ringer's for admixture preparation. Reformulated Zosyn (approved 2005) may provide useful options of drug administration to healthcare professionals to lessen levels of particulates. Supportive data is provided for the expanded compatibility of reformulated Zosyn with different types of Ringer's solutions used globally and for the Y-site coadministration of amikacin and gentamicin aminoglycosides.

Keywords: Zosyn®, aminoglycoside, piperacillin, particulate matter

Introduction

Zosyn® background information

Zosyn®, also known as Tazocin® piperacillin/tazobactam (Wyeth Pharmaceuticals Inc., Philadelphia, PA, USA), is an intravenously administered antibiotic which is composed at an 8:1 ratio of piperacillin (a semisynthetic β-lactam) and tazobactam (a β-lactamase inhibitor derived from the penicillin nucleus). The Food and Drug Administration (FDA) approved it for use in the United States in 1993. Zosyn is currently used in hospitals worldwide to treat patients with moderate-to-severe infections caused by piperacillin-resistant, piperacillin/tazobactam-susceptible β-lactamase-producing strains of certain specified microorganisms. Zosyn is indicated for the treatment of moderate to severe hospital-acquired pneumonia, complicated intra-abdominal infections, complicated skin and soft tissue infections (Wyeth Prescribing Information [Glass vials] 2007; Wyeth Prescribing Information [Galaxy® bags] 2007), and moderate community-acquired pneumonia. Zosyn and Tazocin represent the same product sold by Wyeth in different countries and are used interchangeably in this communication.

History of reformulation of Zosyn

Zosyn, as an injectable antibiotic, is reconstituted and admixed with a variety of diluents before it is administered to a patient. Post-approval, during the 1990's, Zosyn

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had sporadic batch failures related to subvisible particulate matter when conducting the usual Particulate Matter Test (United States Pharmacopoeia [USP] <788>), at the new drug application (NDA) limits of 10,000 (particles ≥10 μM per small volume dose)/1,000 (particles ≥25 μM per small volume dose).

Two key factors identified as responsible for excessive particulates were: (1) small traces of nontoxic silicone oil used to lubricate the elastomeric closures were dispersed in the reconstituted product as an emulsion. The dispersed silicone oil emulsion particles were interpreted by the HIAC particle size analyzer as solid particulate matter, contributing to the out of specification results for sub-visible particulate matter and, (2) subvisible particulate counts increased with the age of the finished product associated with down-shift in the pH of the reconstituted solution of the drug product towards the acidic side.

In response, Wyeth modified its container closure to control silicone lubricant from the rubber stopper insertion process. In addition, the product expiry dating was shortened from 36 to 24 months, as a slight increase occurred in the particulate matter test results in aged product. However, even with these changes, the product still experienced sporadic failures related to particulates. In 1995, USP and European Pharmacopeias adjusted the limits for particulates downwards in part due to ongoing awareness that particulates have the potential to cause morbidity or mortality in patients and because particulate matter had been linked to injection site reactions and thromboembolic events (Longe 1980; Falchuk et al 1985; Lehr et al 2002).

In 2001, partially in response to FDA requests regarding another Wyeth intravenous (IV) product and partially because of FDA concerns about Zosyn, Wyeth undertook to ensure that Zosyn met the revised (1995) USP <788> specifications. The revised specifications lowered the acceptable levels of subvisible particulate matter from 10,000 (particles ≥10 μM per small volume dose)/1,000 (particles ≥25 μM per small volume dose) to 6,000 (particles ≥10 μM per small volume dose)/600 (particles ≥25 μM per small volume dose). This corresponds to a required reduction in the acceptable number of 10 μm and 25 μm particle counts by 40%.

To accomplish this, Wyeth conducted due diligence investigations to establish the root cause and discovered that low pH and trace metal ions can be encountered (Desai et al 2007a, 2007b) through the use of various commercial diluents. Both factors may increase the rate of particulate matter formation in admixtures prepared from Zosyn and potentially penicillins and other β-lactams. This can cause the products to fail USP <788>. The particulate failures in Zosyn may occur even though the pH of the drug substance is in its approved range of 5.5 to 6.8. Wyeth took corrective action by reformulating Zosyn.

Additional limitations of the original formulation and associated stability problems

In 1993, when Zosyn was introduced, its label instructed that the mixing of Zosyn with an aminoglycoside in vitro can result in substantial inactivation of the aminoglycoside. Chemical degradation of the aminoglycoside (possibly involving microbiologically inactive penicillin-aminoglycoside complexes or metal ion leachables) was observed to result in subpotent doses of the aminoglycoside. Such coadministration was also believed to lead to an unacceptably high particulate load (Desai et al 2007a, 2007b). In addition, the label of Zosyn instructed that lactated Ringer's injection solution (LRS) was not compatible with Zosyn and hence could not be used as a reconstitution or admixture diluent.

Transition metal ions as leachables from exogenous/endogenous sources

Contaminants in a drug product solution are defined as "leachables." Potentially contaminating components of the drug product's container system are termed "extractables." Potential links between these two types of compounds have formed much of the basis for the increased scrutiny associated with revisions to the USP-NF and its General Chapter <788> standards for particulate matter in injectable solutions. Jenke (2005) recently proposed a paradigm for the assessment of interaction between these two classes of compounds based upon the origin of and ability to detect extractables in containers, relative to drug substances they may contain. This report cites the example of a regulatory body imposing a requirement of knowledge of such a linkage upon a drug manufacturer, and its potential impact upon a product's life cycle (Jenke 2005).

Reformulation of Zosyn to overcome limitations

Desai and colleagues (2007a, 2007b) recently reported that there is significant inter- and intra-batch variability in both pH and zinc ion content in commercially available IV diluents and solutions across different US manufacturers. Healthcare practitioners administering drugs in these diluents are generally not aware of this variability. In addition, since zinc

is a common transition metal in drug-container packaging and delivery systems, it can leach into the IV solution from container components as well, and inactivate a drug via catalysis (Desai et al 2007a, 2007b).

Commonly used IV diluents may contain small amounts of a variety of exogenous inorganic ions and/or organic substances extracted from the containers and closures into the product during manufacturing or storage. Zinc oxide is used as filler in rubber elastomeric closures of vials, septa at the necks of infusion bags, or in the plungers of infusion syringes. Zinc-based organic substances are also used as polymerization initiators for the polymers used for the construction of admixture bags or infusion line tubing. Zinc and other transition metal ions are known to catalyze chemical degradation of drug molecules (Bandoh et al 1991; Desai et al 2007a, 2007b).

Degradation pathways of antibiotics may include acid hydrolysis, ß-lactam ring opening (Bandoh et al 1991), or epimerization. In the case of ionizable drugs generating anionic species, zinc, calcium, and other metal ions are known to form insoluble solid particulates. In the clinical field, risks associated with the injection of intravenous/intrathecal solutions with particulate contamination are well documented (Nath et al 2004).

The target pH listed on the diluent bags from commercial suppliers can be 3.2 to 6.5 for 5% dextrose injection, and 4.5 to 7.0 for 0.9% sodium chloride injection. The low pH extremes of 3.2 (for 5% dextrose injection) and 4.5 (for 0.9% sodium chloride injection) are considered to be acidic, thus having the potential to accelerate drug degradation, especially in the presence of zinc (Desai et al 2007a, 2007b).

Thus, for the therapeutic performance and safety of drug admixtures infused by the IV route, having data of the pH and levels of zinc in commercial diluents from various parenteral manufacturers is crucial. Since this information is not readily available to the health professionals preparing the solution for parenteral administration, the availability of a drug product resilient to pH or trace metal ion levels commonly found in the clinical setting is preferable.

As part of the root cause analysis, the particulates were isolated and confirmed to be dimers of piperacillin by spectroscopic characterization. The chemical pathway for the dimer formation is shown in Figure 1.

After extensive research, particulate formation in Zosyn was found to be related to the opening of the β-lactam ring of piperacillin, which is accelerated in acidic pH solutions, and is followed by the dimerization of piperacillin molecules that may be further catalyzed by zinc ions.

In summary, the presence of zinc may contribute to incompatibility by: (1) catalysis of drug degradation and potential underdosing, and (2) generation of insoluble solid particulate matter from the dimer formation and/or chelation with ionic species of the drug. Thus, Desai and colleagues (2007a, 2007b)concluded that it is highly desirable that all new drug molecules be screened in the early product

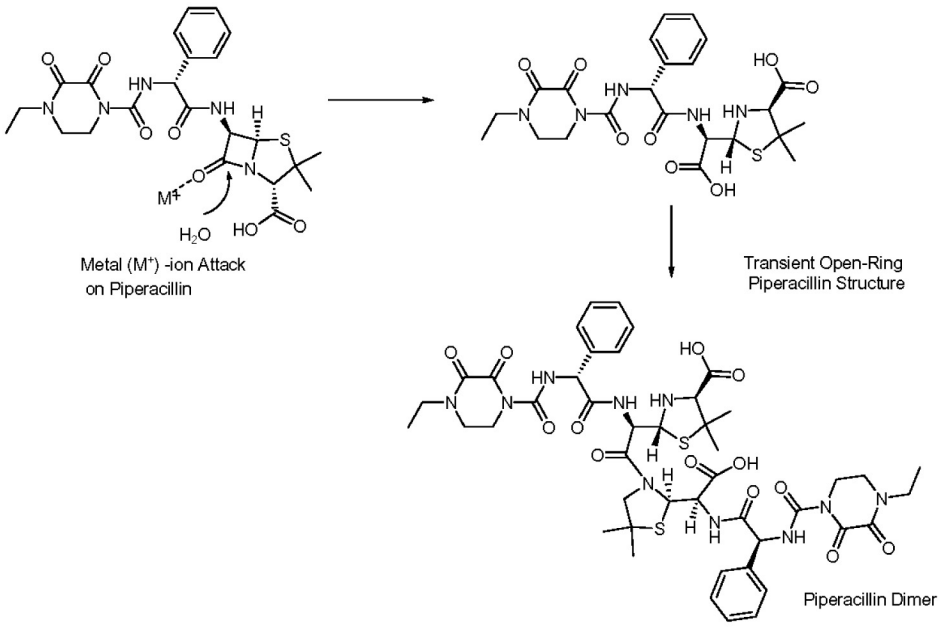

Figure 1 Hydrolysis of piperacillin followed by formation of a piperacillin dimer with low solubility.

development phase for susceptibility to catalysis by zinc, and that the drug product be designed from the outset with sufficient robustness to withstand exposure to variable amounts of zinc ion. It is also recommended that USP monographs address the potential issues posed by zinc in the drug manufacturing and dosage form infusion process.

Combination therapy with Zosyn and amikacin/gentamicin

Because of the prevalent clinical need to administer piperacillin/tazobactam in combination with the aminoglycosides amikacin or gentamicin to treat nosocomial pneumonia caused by *Pseudomonas aeruginosa* as well as conditions caused by other pathogens, the clinician often faces operational challenges: either administer Zosyn and amikacin or gentamicin separately, or through the same intravenous line, typically through a Y-site tube. The latter mode, if possible, would expedite the treatment of the patients in critical care. Mixing of the original formulation of Zosyn with an aminoglycoside in vitro resulted in substantial inactivation of the aminoglycoside by piperacillin. It was postulated that penicillin-aminoglycoside complexes are created that are microbiologically inactive and are of unknown toxicity (Benveniste and Davis 1973; Glew and Pavuk 1983).

Evaluation of reformulated Zosyn to expand compatibility

The compatibility of reformulated Zosyn was evaluated by different physicochemical and spectroscopic methods to show that it can be coadministered as admixture with different types of Ringer's solutions which was not feasible with the original Zosyn. Also, the feasibility of Y-site coadministration with amikacin or gentamicin aminoglycosides was evaluated as shown in the following sections.

Materials and methods

The samples of reformulated Zosyn from Wyeth, used in the admixture diluent compatibility and Y-site aminoglycoside studies, were manufactured and released as per approved product specifications. Commercial intravenous diluents, amikacin and gentamicin antibiotic products were purchased from US/German or other European countries and were used within expiry dating.

For the simulated Y-site administration study, reformulated Zosyn was admixed in a (high dose, low dose) crossover with two different gentamicin drug products in a (high dose, low dose) matrix design. The crossover pairings of reformulated Zosyn and gentamicin were based on the dose extremes expected in clinical use and were derived from the label dosing instructions listed on the package insert for each drug product.

The subvisible particulate (10 and 25 μm) counts were measured by the HIAC light obscuration method described in USP<788> and equivalent EU pharmacopeia 5.0 (2004) 2.9.19 sections.

Typical HPLC assay methods were used for potency determinations of amikacin, piperacillin, and tazobactam in the presence of reformulated Zosyn components.

For gentamicin potency determinations in the presence of reformulated Zosyn components, it was necessary to design a customized nuclear magnetic resonance (NMR) method. For the NMR study, it was necessary to remove the water from the commercial gentamicin product by lyophilization. The simulated Y-site samples were prepared by dissolving lyophilized gentamicin and reformulated Zosyn (lyophilized powder) in D_2O in accordance with the concentration matrix described in Table 1.

Initially, the NMR method by Professor Holzgrabe (Winters 2005) was evaluated to quantify gentamicin potency in the presence of reformulated Zosyn as a function of time. The NMR method appears to be applicable only to

Table 1 Combination matrix for reformulated Tazocin® plus gentamicin

Combination matrix for reformulated Tazocin® plus gentamicin (reconstituted and diluted in D_2O) at clinically relevant bracketed ranges of concentrations

Tazocin sample		Concentration [Pip] and [Tazo]		Gentamicin
	mg/mL in the "bag"[a]	mg/mL in the Y-site sample[b]	mg/mL in the "bag"[a]	mg/mL in the Y-site sample[b]
Reformulated Tazocin High case	40.0 and 5.0	20.0 and 2.5	3.32 High case	1.66
Reformulated Tazocin High case	40.0 and 5.0	20.0 and 2.5	0.7 Low case	0.35
Reformulated Tazocin Low case	13.33 and 1.67	6.67 and 0.84	3.32 High case	1.66
Reformulated Tazocin Low case	13.33 and 1.67	6.67 and 0.84	0.7 Low case	0.35

Notes: [a]The concentrations listed represent the calculated theoretical high and low concentrations that a hospital pharmacist would prepare in separate intravenous infusion bags for reformulated Tazocin and gentamicin; [b]When infused through a Y-site tube, each drug solution will be infused at the same rate. Thus, the resulting admixture solution infused intravenously to the patient will have a final concentration that is half of the concentrations listed above in each individual bag. Therefore, to simulate this practice, the final admixtures will be prepared at concentrations that are half what is listed in the table (eg, For "High Tazocin" – "High Gentamicin" Y-site infusion simulation the sample will be prepared at approximately 20 mg/ml of piperacillin, 2.5 mg/ml of tazobactam, and 1.66 mg/ml of gentamicin in D_2O.).

gentamicin drug product as-is, with a low ambient pH of about 4 to 4.6. Upon the addition of reformulated Zosyn, the final pH of the simulated Y-site solution with gentamicin is raised to about 6, so the ionic environment of the anomeric protons and the –N-methyl protons is changed, and hence the NMR spectrum is totally different than that of gentamicin drug product alone as observed by the Holzgrabe group.

For the customized NMR method, two groups of proton resonances of gentamicin were isolated from other proton resonances and were explored for the quantification of gentamicin. One is from the anomeric protons at δ_H 5.84 (doublet) and δ_H 5.80 (multiplet). The other is from the N-methyl group at δ_H 2.85 (singlet). Although the anomeric proton resonances were well separated, due to the relatively low concentration of gentamicin used in the clinical admixtures, the observed signal-to-noise ratio for the anomeric protons was very low. The N-methyl proton resonance peaks (with very good separation from the other peaks, and a peak intensity significantly higher than the anomeric proton peaks) were considered more appropriate to provide quantification of clinically relevant parenteral admixtures with low levels of gentamicin in the presence of very high levels of reformulated Zosyn.

NMR experiments were performed at room temperature on a Bruker DRX-500 NMR spectrometer, operating at 500.13 MHz (^{1}H), equipped with TOPSPIN software (Version 1.3). 112 scans were collected into 64 k data points giving a digital resolution of 0.16 Hz per point. The spectral width was 10330 Hz, the transmitter offset at 6.17 ppm, and the flip angle was 90°. Using an acquisition time of 3.17 s and an additional delay of 1 s, the pulse repetition period was about 4.17 s. Samples were measured in D_2O at 298 K. Each ^{1}H NMR data point for gentamicin represents cumulative scans collected over ten minutes.

Hydroquinone monoethyl ether was used as an internal standard due to the absence of chemical interactions with gentamicin and all components of reformulated Zosyn. Also, the associated peaks of the hydroquinone monoethyl ether protons do not overlap or cause interference with the N-methyl protons of gentamicin that were used for quantification. This customized NMR method provides a kinetic snapshot at 10-minute intervals over a period of 1 hour for gentamicin strength in the presence of reformulated Zosyn.

Results and discussion

Improved clinical utility of the reformulated version of Zosyn

In 2005 (USP <788> 2007), the FDA approved the new formulation of Zosyn that complies with USP<788> particulate specifications and has an expanded compatibility profile with the aminoglycosides, amikacin and gentamicin. The modifications to the formulation consisted of the addition of the disodium salt of ethylene diamine tetraacetic acid (disodium EDTA; edetate disodium dihydrate), which acts as a metal-chelating agent, and sodium citrate, which acts as a buffer. These modifications lessen the possibility of particulate matter accumulation during storage of the solution form of Zosyn or the development of particulate matter upon reconstitution of Zosyn lyophilized powder with commonly used diluents. In addition, reformulated Zosyn can be administered simultaneously, either with amikacin or gentamicin, via Y-site infusion at specific doses and concentrations, and with certain diluents (Wyeth Prescribing Information [Glass vials] 2007a; Wyeth Prescribing Information [Galaxy® bags] 2007b).

Also, unlike original Zosyn, reformulated Zosyn has been shown (Table 2) to be compatible with LRS or Hartmann's solution (European version of LRS) for dilution. Healthcare practitioners sometimes prefer to use LRS or Hartmann's solution.

Allowance of Y-site coadministration of reformulated Zosyn with aminoglycosides

The inactivation of aminoglycosides in the presence of penicillin-class drugs containing β-lactam rings has been recognized (Benveniste and Davis 1973; Glew and Pavuk 1983). However, amikacin and gentamicin have been shown to be compatible in vitro with reformulated Zosyn containing disodium EDTA supplied in vials or bulk pharmacy containers in certain diluents at specific doses and concentrations for a simultaneous Y-site infusion.

Simulated Y-site administration studies by Wyeth were conducted for reformulated Zosyn with amikacin or gentamicin in the concentration ranges and clinical doses described in the labels of Zosyn and each of the aminoglycosides. The simulated studies mimicked Y-site coadministration for Zosyn-amikacin systems and evaluated potency and degradation products by HPLC analyses (Tables 3 and 4). Reformulated Zosyn was shown to be compatible for simultaneous administration via a Y-site intravenous tube with amikacin in the concentration ranges of 2.25 g reformulated Zosyn/150 mL to 4.5 g/50 mL for Zosyn and 1.75 mg/mL to 7.5 mg/mL for amikacin in sterile water for injection, USP and 0.9% sodium chloride injection, USP, 5% dextrose in water for injection, USP and lactated Ringer's injection, USP (Table 5). Degradation products and related compounds in the solution were quantified in a further study, which confirmed the first study's conclusions, and

Table 2 Summary of drug potency results for admixtures of reformulated Zosyn® in various diluents and stored at room temperature for up to 24 hours

Diluent	Time point (Hours)	Theoretical values (mg/mL)		Observed values (mg/mL)				
		[PIP]	[TAZO]	[PIP]	% Initial PIP	[TAZO]	% Initial TAZO	pH
Saline	0	80.0	10.0	79.91	100.0	10.05	100.0	6.21
Saline	24	80.0	10.0	79.14	99.0	9.90	98.5	6.04
WFI	0	80.0	10.0	77.28	100.0	9.72	100.0	6.48
WFI	24	80.0	10.0	79.26	102.6	9.94	102.2	6.24
D5W	0	80.0	10.0	79.17	100.0	9.95	100.0	6.43
D5W	24	80.0	10.0	77.96	98.5	9.75	98.0	6.23
Compound sodium lactate intravenous infusion BP (Hartmann's solution)	0	16.0	2.0	15.32	100.0	1.93	100.0	6.10
Compound sodium lactate intravenous infusion BP (Hartmann's solution)	24	16.0	2.0	14.68	95.8	1.84	95.3	5.92
Saline	0	13.33	1.67	13.79	100.0	1.71	100.0	6.30
Saline	24	13.33	1.67	13.92	100.9	1.72	100.6	6.03
WFI	0	40.0	5.0	38.16	100.0	4.75	100.0	6.53
WFI	24	40.0	5.0	38.83	101.8	4.80	101.1	6.33
D5W	0	13.33	1.67	13.16	100.0	1.63	100.0	6.59
D5W	24	13.33	1.67	13.69	104.0	1.69	103.7	6.31
Compound sodium lactate intravenous infusion BP (Hartmann's solution)	0	8.0	1.0	8.32	100.0	1.03	100.0	6.09
Compound sodium lactate intravenous infusion BP (Hartmann's solution)	24	8.0	1.0	8.25	99.2	1.02	99.0	5.90

Abbreviations: PIP, Piperacillin; Saline, 0.9% NaCl solution; TAZO, Tazobactam; WFI, water for injection; D5W, 5% dextrose solution.

Table 3 Summary of drug potency results for simulated Y-site coadministration of reformulated Zosyn® with amikacin in compound sodium lactate intravenous infusion BP (Hartmann's solution) at room temperature

Time point (Hours)	Theoretical values (mg/mL)			Observed potency (mg/mL) and % Remaining of three Antibiotics						
	[Pip]	[Tazo]	[Amik]	[Pip]	% Initial Pip	[Tazo]	% Initial Tazo	[Amik]	% Initial Amik	pH
0	8.0	1.0	3.75	8.03	100.0	1.00	100.0	3.75	100.0	5.39
1	8.0	1.0	3.75	8.02	99.9	1.00	100.0	3.75	100.0	5.36
2	8.0	1.0	3.75	8.04	100.1	1.00	100.0	3.74	100.0	5.36
4	8.0	1.0	3.75	8.02	99.9	1.00	100.0	3.77	100.5	5.35
0	8.0	1.0	0.875	8.18	100.0	1.02	100.0	0.88	100.0	5.65
1	8.0	1.0	0.875	8.06	98.5	1.00	98.0	0.88	100.0	5.64
2	8.0	1.0	0.875	8.13	99.4	1.01	99.0	0.89	101.1	5.66
4	8.0	1.0	0.875	8.18	100.0	1.02	100.0	0.89	101.1	5.65
0	4.0	0.5	3.75	4.19	100.0	0.52	100.0	3.76	100.0	5.31
1	4.0	0.5	3.75	4.19	100.0	0.52	100.0	3.76	100.0	5.30
2	4.0	0.5	3.75	4.22	100.7	0.52	100.0	3.77	100.2	5.30
4	4.0	0.5	3.75	4.15	99.0	0.51	98.1	3.75	99.7	5.29
0	4.0	0.5	0.875	4.35	100.0	0.53	100.0	0.88	100.0	5.60
1	4.0	0.5	0.875	4.35	100.0	0.54	101.9	0.88	100.0	5.59
2	4.0	0.5	0.875	4.36	100.2	0.54	101.9	0.89	101.1	5.60
4	4.0	0.5	0.875	4.28	98.4	0.53	100.0	0.89	101.1	5.59

Note: The same intravenous diluent was used to reconstitute the lyophilized drug and to prepare the intravenous infusion at the proper concentration for administration to the patient. In this case it was compound sodium lactate intravenous infusion BP (Hartmann's solution).
Abbreviations: Amik, amikacin; Pip, piperacillin; Tazo, Tazobactam.

Table 4 Degradation products of reformulated Zosyn® drug components in the presence of amikacin up to 4 hours at room temperature in compound sodium lactate intravenous infusion BP (Hartmann's solution)

Time point (Hours)	Theoretical potency (mg/mL)		% of degradation products[a] of Zosyn® drug components				
			RRT 0.104	RRT 0.516	RRT 0.564	RRT 0.591	RRT 0.635
	[Pip]/[Tazo]	[Amik]					
0	8.0/1.0	3.75	0.056	0.59	–	0.078	–
1	8.0/1.0	3.75	0.067	0.72	–	0.078	–
2	8.0/1.0	3.75	0.067	0.89	–	0.078	–
4	8.0/1.0	3.75	0.078	1.13	–	0.067	–
0	8.0/1.0	0.875	0.067	0.56	–	0.078	–
1	8.0/1.0	0.875	0.067	0.58	–	0.078	–
2	8.0/1.0	0.875	0.067	0.62	–	0.078	–
4	8.0/1.0	0.875	0.067	0.70	–	0.078	–
0	4.0/0.5	3.75	0.067	0.56	0.10	0.089	0.20
1	4.0/0.5	3.75	0.067	0.76	–	0.089	0.17
2	4.0/0.5	3.75	0.067	0.89	–	0.089	0.24
4	4.0/0.5	3.75	0.089	1.18	–	0.067	0.10
0	4.0/0.5	0.875	0.067	0.56	–	0.089	0.10
1	4.0/0.5	0.875	0.067	0.60	–	0.089	0.10
2	4.0/0.5	0.875	0.067	0.62	–	0.089	0.11
4	4.0/0.5	0.875	0.067	0.69	–	0.089	0.08

Abbreviations: RRT, Relative retention times of the HPLC chromatograms corresponding to the known degradation products of Zosyn® and amikacin in simulated y-site mixtures.

Notes: [a]The observed % value of the degradation products are far below the approved specifications for the products.

showed that reformulated Zosyn (piperacillin/tazobactam) is compatible for Y-site coadministration with amikacin or gentamicin. US-sourced amikacin and gentamicin, evaluated here, represent the composition ranges of the products prescribed globally.

The study designs used for Y-site compatibility and admixture stability are based on simulated Y-site injection testing procedures reported by Choi and colleagues (1994) and by Trissel and Martinez (1994a).

A follow-up simulated Y-site compatibility study, conducted by using an innovative NMR method, is supplementary to the previous study using an LC-MS method. The current study differs from the previous study in that it tested the simulated Y-site mixtures of reformulated Tazocin

Table 5 Simulated Y-site coadministration of reformulated Zosyn® with amikacin in different admixture diluents (Potency of antibiotics at 4 hours)

Admixture diluent	Initial composition		% Antibiotic remaining of the initial		
	Zosyn® (PIP mg/mL/ Tazo mg/mL)	Amikacin (mg/mL)	% Amikacin	% Tazobactam	% Piperacillin
0.9% NaCl	80/10	7.5	95.7	98.6	98.7
0.9% NaCl	80/10	1.75	105.7	99.0	99.3
0.9% NaCl	13/1.7	7.5	99.8	98.5	98.8
0.9% NaCl	13/1.7	1.75	111.0	99.5	99.4
5% Dextrose	80/10	7.5	94.0	104.9	105.3
5% Dextrose	80/10	1.75	90.0	99.7	99.9
5% Dextrose	13/1.7	7.5	97.6	99.4	99.4
5% Dextrose	13/1.7	1.75	94.4	98.9	99.1
WFI/ 0.9% NaCl	80/10	7.5	99.2	98.6	98.9
WFI/ 0.9% NaCl	80/10	1.75	97.5	101.1	99.3
WFI/ 0.9% NaCl	13/1.7	7.5	100.3	96.2	97.8
WFI/ 0.9% NaCl	13/1.7	1.75	100.3	98.3	98.5
Lactated Ringers	16/2	7.5	93.3	99.0	98.3
Lactated Ringers	16/2	1.75	92.6	99.8	99.4
Lactated Ringers	8/1	7.5	99.7	98.6	99.0
Lactated Ringers	8/1	1.75	97.3	99.6	99.6

Abbreviations: WFI, water for injection; PIP, piperacillin; Tazo, Tazobactam.

and gentamicin at shorter time intervals of 0, 10, 20, 30, 40, 50, and 60 minutes, while the previous study tested these mixtures at 0, 1, 2, and 4 hours. This change is to collect data at clinically realistic shorter time intervals, because the 4-hour testing time point represents an exaggerated contact time for the Y-site drug compatibility testing. In actual clinical practice, when two drugs are infused through a Y-site the maximum estimated contact time (prior to entering the blood stream) is short, often in the range of 15 minutes and not extending more than 60 minutes (Trissel 1994b; Leissing 1989). In this experiment to simulate Y-site administration, reformulated Tazocin was admixed in a (high dose, low dose) crossover with two gentamicin drug products sourced from Germany in a (high dose, low dose) matrix design, as shown in Table 1. The crossover pairings of reformulated Tazocin and gentamicin were based on the dose extremes expected in clinical use and were derived from the label dosing instructions listed on the package insert for each drug product.

Simulated Y-site stability results

The kinetic snapshot at every ten minutes over a period of 0 to 60 minutes of the mixed solutions analyzed by evaluating the –N-methyl proton resonance of the gentamicin molecule in a noninvasive manner at room temperature shows the following:

(1) For both gentamicin products in all four combinations with reformulated Tazocin, the potency values of gentamicin at ten minutes are maintained at greater than 98% of the initial. As described earlier, the typical time for gentamicin and reformulated Tazocin to remain in contact during the actual Y-site co-administration in a clinical setting is about 15 minutes.

(2) At 30 minutes and 60 minutes, better than 97% and 96% of the initial potency of gentamicin is maintained, respectively, even for the worst-case scenario of a solution mixture of "high" reformulated Tazocin and "low" gentamicin during a simulated Y-site administration. Based on the aminoglycoside and β-lactam interaction chemistry, the highest degradation of gentamicin was expected for this combination.

(3) As expected, for the "high" gentamicin combination with "low" or "high" reformulated Tazocin, the gentamicin strength at 60 minutes was found to be greater than 98%.

Representative data for Y-site compatibility at clinically relevant concentration ranges of reformulated Tazocin combined with another German gentamicin commercial drug product (Ratiopharm®) are provided in Table 6. Data

Table 6 Reformulated Tazocin®: Simulated Y-site compatibility by NMR for Ratiopharm® German gentamicin drug product

Product Name	Gentamicin - Ratiopharm® 40 SF (40 mg/1 ml ampoule gentamicin, Ratiopharm® Company, Germany)							
Components	Gentamicin sulfate Acetylcysteine Na_2EDTA NAOH or H_2SO4 for pH adjustment WFI							
Combination matrix in simulated y-site Prepared in D_2O solution	High Tazo: High Genta (20 mg/mL Pip: 1.6 mg/mL Genta)[a]		High Tazo: Low Genta (20 mg/mL Pip: 0.35 mg/mL Genta)[a]		Low Tazo: High Genta (6.67 mg/mL Pip: 1.6 mg/mL Genta)[b]		Low Tazo: Low Genta (6.67 mg/mL Pip: 0.35 mg/mL Genta)[b]	
Sample number	L39609-28-1		L39609-28-2		L39609-28-3		L39609-28-4	
Time (min)	Relative peak area δ_H:2.85 ($-NCH_3$)[c]	% Initial[d]	Relative peak area δ_H:2.85 ($-NCH_3$)[c]	% Initial[d]	Relative peak area δ_H:2.85 ($-NCH_3$)[c]	% Initial[d]	Relative peak area δ_H:2.85 ($-NCH_3$)[c]	% Initial[d]
0	1.7558	100	0.3942	100	1.8981	100	0.4194	100
10	1.7533	99.9	0.3881	98.5	1.8948	99.8	0.4170	99.4
20	1.7518	99.8	0.3846	97.6	1.8953	99.9	0.4183	99.7
30	1.7512	99.7	0.3847	97.6	1.8971	99.9	0.4131	98.5
40	1.7450	99.3	0.3792	96.2	1.8977	100.0	0.4135	98.6
50	1.7496	99.6	0.3784	96.0	1.8897	99.6	0.4116	98.1
60	1.7453	99.4	0.3806	96.5	1.8929	99.7	0.4102	97.8

Notes: [a]also contains 2.5 mg/ml of tazobactam; [b]also contains 0.84 mg/ml of tazobactam; [c]observed peak area for δ_H: 2.85 ($-NCH_3$) of gentamicin is relative to internal standard of hydroquinone methyl ether; [d]% of initial concentration of gentamicin remaining.
Abbreviations: Tazo: Tazocin®; Pip, piperacillin.

Table 7 Summary of subvisible particulate counts by HIAC for simulated Y-site coadministration of reformulated Zosyn® and amikacin

Sample description	Reconstitution solvent	Solution concentration	Aminoglycoside concentration	Admixture diluent	Particulate counts by HIAC			
					10 μm t = 0 hrs	10 μm t = 4 hrs	25 μm t = 0 hrs	25 μm t = 4 hrs
Particulate Count Specifications as per USP <788>[a]					**NMT 6000**	**NMT 6000**	**NMT 600**	**NMT 600**
Low Zosyn	WFI	40 mg/mL piperacillin plus 5 mg/mL tazobactam	Amikacin (7.5 mg/ml)	Saline	2595 2920 1875	1585 1320 3640	45 60 25	45 40 30
Low Zosyn	WFI	40 mg/mL piperacillin plus 5 mg/mL tazobactam	Amikacin (1.75 mg/ml)	Saline	4505 4265 4555	1265 1465 1450	70 35 80	0 20 25
High Zosyn	WFI	80 mg/mL piperacillin plus 10 mg/mL tazobactam	Amikacin (7.5 mg/ml)	Saline	4430 4270 2235	750 2290 3320	100 50 45	15 10 40
High Zosyn	WFI	80 mg/mL piperacillin plus 10 mg/mL tazobactam	Amikacin (1.75 mg/ml)	Saline	3780 4840 3965	1160 455 1255	50 105 25	35 0 35
Low Zosyn	Saline	13.33 mg/mL piperacillin plus 1.67 mg/mL tazobactam	Amikacin (7.5 mg/ml)	Saline	845 1210 1600	1730 815 1250	15 15 20	35 65 45
Low Zosyn	Saline	13.33 mg/mL piperacillin plus 1.67 mg/mL tazobactam	Amikacin (1.75 mg/ml)	Saline	555 1670 1940	2150 1890 1485	0 15 10	120 75 115
High Zosyn	Saline	80 mg/mL piperacillin plus 10 mg/mL tazobactam	Amikacin (7.5 mg/ml)	Saline	1015 3145 2320	5690 1295 5360	15 0 50	125 30 160
High Zosyn	Saline	80 mg/mL piperacillin plus 10 mg/mL tazobactam	Amikacin (1.75 mg/ml)	Saline	3960 1765 2150	1665 3225 1635	25 20 15	40 75 100
Low Zosyn	Lactated Ringer's	8 mg/mL piperacillin plus 1 mg/mL tazobactam	Amikacin (7.5 mg/ml)	Lactated Ringer's	1180 915 640	185 285 580	5 0 5	0 0 0
Low Zosyn	Lactated Ringer's	8 mg/mL piperacillin plus 1 mg/mL tazobactam	Amikacin (1.75 mg/ml)	Lactated Ringer's	505 490 815	285 365 170	15 5 0	0 5 10
High Zosyn	Lactated Ringer's	16 mg/mL piperacillin plus 2 mg/mL tazobactam	Amikacin (7.5 mg/ml)	Lactated Ringer's	645 635 1245	415 380 275	0 15 20	20 0 0
High Zosyn	Lactated Ringer's	16 mg/mL piperacillin plus 2 mg/mL tazobactam	Amikacin (1.75 mg/ml)	Lactated Ringer's	600 495 385	275 505 415	15 10 5	5 10 10

Abbreviations: NMT, not more than; WFI, water for injection; Saline, 0.9% sodium chloride.
Notes: [a]Particulate count specifications in EU and other Pharmacopeia are similar.

Table 8 Summary of subvisible particulate counts by HIAC for simulated Y-site coadministration of reformulated Zosyn® and gentamicin

Sample description	Reconstitution solvent	Solution concentration	Aminoglycoside concentration	Admixture diluent	Particulate counts by HIAC			
					10 μm t = 0 hrs	10 μm t = 4 hrs	25 μm t = 0 hrs	25 μm t = 4 hrs
Particulate Count Specifications as per USP <788>					**NMT 6000**	**NMT 6000**	**NMT 600**	**NMT 600**
Low Zosyn	WFI	40 mg/mL piperacillin plus 5 mg/mL tazobactam	Gentamicin (3.32 mg/ml)	Saline	545 435 570	1750 715 1555	20 5 15	5 10 15
Low Zosyn	WFI	40 mg/mL piperacillin plus 5 mg/mL tazobactam	Gentamicin (0.7 mg/ml)	Saline	550 1745 730	1435 390 395	5 5 10	50 5 15
High Zosyn	WFI	80 mg/mL piperacillin plus 10 mg/mL tazobactam	Gentamicin (3.32 mg/ml)	Saline	1470 1980 540	110 170 1110	35 10 0	0 25 60
High Zosyn	WFI	80 mg/mL piperacillin plus 10 mg/mL tazobactam	Gentamicin (0.7 mg/ml)	Saline	1170 2770 1465	600 310 210	30 20 20	50 5 20
Low Zosyn	Saline	13.33 mg/mL piperacillin plus 1.67 mg/mL tazobactam	Gentamicin (3.32 mg/ml)	Saline	765 4220 2700	1465 1130 1485	5 145 35	70 30 50
Low Zosyn	Saline	13.33 mg/mL piperacillin plus 1.67 mg/mL tazobactam	Gentamicin (0.7 mg/ml)	Saline	935 1500 1375	4425 2465 760	25 50 25	70 30 50
High Zosyn	Saline	80 mg/mL piperacillin plus 10 mg/mL tazobactam	Gentamicin (3.32 mg/ml)	Saline	3385 2275 2265	4120 2560 2660	140 35 60	140 110 120
High Zosyn	Saline	80 mg/mL piperacillin plus 10 mg/mL tazobactam	Gentamicin (0.7 mg/ml)	Saline	2035 5235 2790	1175 1865 1800	45 215 80	45 105 60
Low Zosyn	5% Dextrose	13.33 mg/mL piperacillin plus 1.67 mg/mL tazobactam	Gentamicin (3.32 mg/ml)	5% Dextrose	860 1175 695	540 435 445	25 25 20	165 75 95
Low Zosyn	5% Dextrose	13.33 mg/mL piperacillin plus 1.67 mg/mL tazobactam	Gentamicin (0.7 mg/ml)	5% Dextrose	515 310 505	190 165 305	20 5 10	15 20 15
High Zosyn	5% Dextrose	80 mg/mL piperacillin plus 10 mg/mL tazobactam	Gentamicin (3.32 mg/ml)	5% Dextrose	2545 1905 1030	675 845 985	65 55 40	20 20 20
High Zosyn	5% Dextrose	80 mg/mL piperacillin plus 10 mg/mL tazobactam	Gentamicin (0.7 mg/ml)	5% Dextrose	1860 2825 2240	690 680 630	25 15 10	20 5 25
Low Zosyn	Lactated Ringer's	8 mg/mL piperacillin plus 1 mg/mL tazobactam	Gentamicin (3.32 mg/ml)	Lactated Ringer's	1120 930 135	170 85 165	0 15 5	5 0 0

(*Continued*)

Table 8 (*Continued*)

Sample description	Reconstitution solvent	Solution concentration	Aminoglycoside concentration	Admixture diluent	Particulate counts by HIAC			
					10 μm t = 0 hrs	10 μm t = 4 hrs	25 μm t = 0 hrs	25 μm t = 4 hrs
Particulate Count Specifications as per USP <788>					**NMT 6000**	**NMT 6000**	**NMT 600**	**NMT 600**
Low Zosyn	Lactated Ringer's	8 mg/mL piperacillin plus 1 mg/mL tazobactam	Gentamicin (0.7 mg/ml)	Lactated Ringer's	1215 250 465	180 325 190	10 5 15	5 10 0
High Zosyn	Lactated Ringer's	16 mg/mL piperacillin plus 2 mg/mL tazobactam	Gentamicin (3.32 mg/ml)	Lactated Ringer's	325 870 1235	270 155 250	5 10 0	0 5 0
High Zosyn	Lactated Ringer's	16 mg/mL piperacillin plus 2 mg/mL tazobactam	Gentamicin (0.7 mg/ml)	Lactated Ringer's	375 410 630	330 205 270	15 5 5	0 0 5

Abbreviation: NMT, not more than.

for the Rebofacin gentamicin product (not shown here) were found to be similar.

Particulate matter evaluation for reformulated Zosyn – amikacin or gentamicin (simulated Y-site study)

The subvisible particle counts for 10 μm and 25 μm size were determined using the HIAC method. The particulate counts at 0 and 4 hours for simulated Y-site mixtures of reformulated Zosyn and gentamicin or amikacin in admixtures of common commercial diluents are provided in Tables 7 and 8. The concentrations of reformulated Zosyn and gentamicin or amikacin were chosen to simulate clinically relevant dilutions in the admixture infused. The particulate counts up to the 4 hour test period for the reformulated Zosyn – amikacin system and reformulated Zosyn – gentamicin system were well within the current USP and EU Pharmacopeia specifications. Reformulated Zosyn was shown to be compatible for coadministration via a Y-site intravenous tube with gentamicin under the concentration ranges of 2.25 g reformulated Zosyn/150 mL to 4.5 g/100 mL for Zosyn and 0.7 mg/mL to 3.32 mg/mL for gentamicin in the 0.9% sodium chloride injection USP coadministration of Zosyn with 0.9% sodium chloride.

Conclusions

In summary, the product quality enhancement provides expanded flexibility for the administration of reformulated Zosyn under variable clinical use conditions:

(1) Reformulated Zosyn complies with USP-NF 30 <788>, European and Asia Pacific Pharmacopoeia specifications for particulate matter in injections under all clinical use conditions because it is tolerant to variability in pH and metal ion concentrations of commercial solutions used in the clinical setting. The variabilities of actual pH and metal ion concentrations in commercial IV solutions and diluents are unknown to the pharmacist and nurse end-user.

(2) The product maintains chemical and physical stability under the conditions encountered in the clinical field of use where commercial diluents, such as 5% dextrose solution with potential variables of pH and leachable metal ions, are used for admixture preparation for parenteral administration. Regardless of the pH or zinc content of an admixture diluent or IV solution, reformulated Zosyn will maintain potency and lessen the level of particulates infused into patients.

(3) The product is compatible with calcium-containing Ringer's solutions (as lactate or acetate) or Hartmann's solution.

(4) The reformulated product provides the capability of simultaneous Y-site coadministration of amikacin (with 0.9% NaCl or 5% dextrose) and gentamicin (with 0.9% NaCl), without compromising either drug's potency, and provides useful options for the administration of Zosyn, especially for the treatment of nosocomially-acquired pneumonia.

(5) Based on the recent stability data, the expiration dating of reformulated Zosyn has been extended to 36 months from its previously reduced dating of 24 months.

References

Bandoh K, Katoh M, Muto Y, et al. 1991. Metal induced degradation of β-lactams. *Chemotherapy*, 39:315–8.

Benveniste R, Davis J. 1973. Structure activity relationships among the aminoglycoside antibiotics: role of hydroxyl and amino groups. *Antimicrob Agents Chemother*, 4:402–9.

Choi JS, Burm JP, Jhee SS, et al. 1994. Stability of piperacillin sodium-tazobactam sodium and ranitidine hydrochloride in 0.9% sodium chloride injection during simulated Y-site administration. *Am J Hosp Pharm*, 51:2273–6.

Desai NR, Vencl-Joncic M, Koczone K, et al. 2007a. Screening of Zinc in commercial intravenous diluents commonly used for reconstitution and admixing of injectable dosage forms, and flushing of administration devices [poster]. *Am Pharm Assoc Ann Meet (Atlanta, GA)*, 167.

Desai NR, Shah SM, Koczone K, et al. 2007b. Zinc content of commercial diluents widely used in drug admixtures prepared for intravenous infusion. *Int J Pharma Compound*, 11:4226–432.

European Pharmacopeia. 2004. Particulate contamination: Subvisible particles. 5.0(2004).2.9.19, pp. 253–5.

Falchuk KH, Peterson L, McNeal J. 1985. Microparticulate induced phlebitis: Its prevention by in-line filtration. *N Eng J Med*, 312:78–82.

Glew RH, Pavuk R. 1983. Stability of gentamicin, tobramicin and amikacin in combination with four β-lactam antibiotics. *Antimicrob Agents Chemother*, 24:474–7.

Jenke DR. 2005. Linking extractables and leachables in container/closure applications. *DA J Pharm Sci Technol*, 59:265–81.

Lehr H-A, Brunner J, Rangoonwala Ret al. 2002. Particulate matter contamination of intravenous antibiotics aggravates loss of functional capillary density in post ischemic striated muscle. *Am J Respir Crit Care Med*, 165:514–20.

Leissing NC, Story KO, Zaske D. 1989. Inline fluid dynamics in piggyback and manifold drug delivery systems, *Am J Hosp Pharm*, 46:89–97.

Longe RL. 1980. Particulate contamination in selected parenteral drugs. *Can Anesthesic Soc J*, 27:62–4.

Nath N, McNeal E, Obenhuber D, et al. 2004. Particulate contaminants of intravenous medication and the limits set by USP General Chapter <788>. *Pharmacopeial Forum*, 30:2272–80.

Trissel LA, Martinez JF. 1994a. Compatibility of piperacillin sodium plus tazobactam with selected drugs during simulated Y-site injection. *Am J Hosp Pharm*, 51:672–8.

Trissel LA, 1994b. Handbook of Injectable Drugs, PP XVIII, 5th Edition, American Society of Health-System Pharmacists® Bethesda, MD, USA.

[USP] United States Pharmacopeia. 2007. USP 30, Supplement 2. <788>, Particulate matter in injections.

Winter W, Deubner R, Holzgrabe U. 2005. Multivariate analysis of nuclear magnetic resonance data-characterization of critical drug substance quality of gentamicin sulfate. *J Pharm Biomed Anal.* 38:833–9.

Wyeth Pharmaceuticals. 2007a. Zosyn® (Piperacillin/Tazobactam For Injection USP [Glass Vials]) United States Prescribing Information.

Wyeth Pharmaceuticals. 2007b. Zosyn® (Piperacillin/Tazobactam For Injection USP [Galaxy® Bags]) United States Prescribing Information.