

# An Improved Procedure for Preparation and Isolation of Cephalosporin Antibiotic: Cefozopran as Free Base

Neera Tewari,\* Bishwa Prakash Rai, Kallimulla Mohammad, Hashim Nizar, and Mohan Prasad

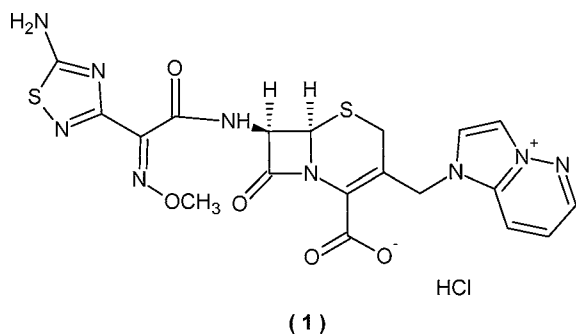
Chemical Research Division, Ranbaxy Research Laboratories, Gurgaon, Haryana 122001, India

## Abstract:

An efficient synthesis of cephalosporin antibiotic, cefozopran is described. The present process does not involve chromatographic purification for isolation and is cost-effective and amenable to large-scale synthesis.

## Introduction

Cefozopran (**1**) is a parenteral broad spectrum fourth-generation cephalosporin having high antibacterial activity against both Gram negative and Gram positive bacteria. It has been clinically available in Japan since the late 1990s for the treatment of various infections such as pneumonia, sepsis, urinary-tract infections, and intraabdominal infections.<sup>1–3</sup>



In the course of our ongoing project on the synthesis of cefozopran (**1**), we have developed an industrially viable process for the preparation of cefozopran free base **4** which is converted to its hydrochloride salt as per conventional methods. Literature survey reveals that **4** is prepared by condensation of 2-(5-amino-1,2,4-thiadiazol-3-yl)-2-(Z)-methoxy iminoacetyl chloride hydrochloride (**2a**) with 7-amino-3-[(imidazo(1,2-*b*)pyridazinium-1-yl)methyl]-3-cephem-4-carboxylate hydroiodide (**3**) in presence of base.<sup>4</sup> (Scheme 1) This method involves condensation at low temperature, isolation using column chromatography followed by lyophilization to afford **4** as an amorphous material. Another method reports<sup>5</sup> the condensation of **3** with (*S*)-(2-benzothiazolyl)-(Z)-2-(2-amino-1,2,4-thiadiazol-3-yl)-2-methoxyiminothioacetate (**2b**) in the presence of tri-*n*-butyl amine

(TBA) in methanol at 0–5 °C, and the product is isolated by column chromatography followed by crystallization at 6–8 °C for 4–6 h to give **4** in 70% yield. The drawback of the first method is the use of column chromatography and lyophilization which provides **4** as an amorphous material that has stability issues. The other method also involves column chromatography for the isolation of crystalline **4**. Both the methods involve exhaustive workup and capital investment. In this communication we report a simplified process over the existing process on commercial scale with improved yields from **2b** and **3**.

## Results and Discussion

The present method describes the synthesis of **4** in one step by the reaction of **2b** with **3** in a mixture of water and THF in the presence of base. The product is directly isolated from the reaction mixture by filtration. In a typical experiment, reactant **2b** and **3** are suspended in a mixture of THF/water (10:5, volume with respect to input of **3**) followed by the addition of tri-*n*-butyl amine at RT (20–30 °C). As the reaction proceeds, the compound **4** separates as a solid, which is filtered and isolated in 91% yield. During our optimization studies we have carried out the reactions under different conditions, and the results are tabulated in Table 1. It is found that varying the ratio of THF/water and base impacts the yield of **4**. The best result is obtained in THF/water (15:8) and TBA as base (entry 3). The advantages of this method are its simplicity in terms of operation on commercial scale and direct isolation of crystalline **4**, thereby eliminating exhaustive workup and column chromatography.

In conclusion, a simplified, cost-effective industrial process for synthesis and isolation of **4** with improved yield is described. This process does not involve any chromatographic separation and purification steps.

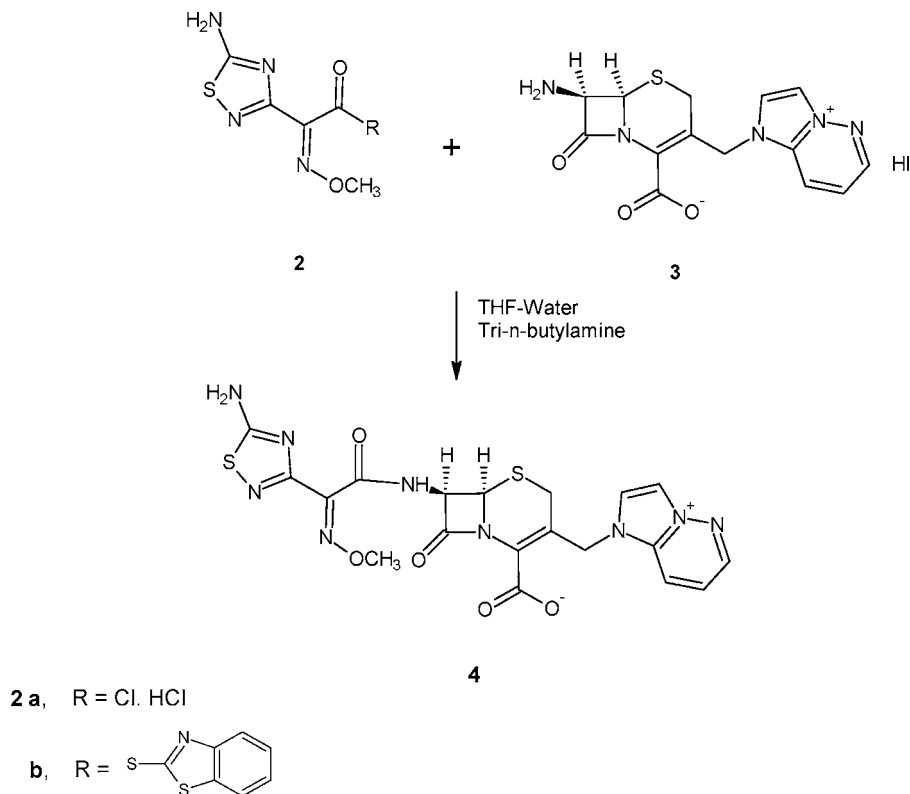
## Experimental Section

**General.** Reagents are used as such without purification. <sup>1</sup>H NMR spectra are recorded using a Bruker 300 MHz spectrometer. The chemical shift data are reported as δ (ppm) downfield from tetramethylsilane which is used as an internal standard. HPLC analysis is performed on a Waters instrument with a UV detector (235 nm) using an Intersil ODS 3 V (250 mm × 4.6 mm, 5 μm) and mobile phase {acetonitrile/buffer (0.02 M sodium dihydrogen orthophosphate dihydrate in water, pH 6.5, adjusted with NaOH solution), 0:100 in 0 min to 90:10 in 12 min; 12 to 40 min 30:70, 40 to 42 min 0:100, 42 to 60 min 0:100} with a flow rate of 1.0 mL/min. Mass spectrum is recorded using an API 2000 (MPS SCIEX) instrument.

\* Author for correspondence. E-mail: neera.tiwari@ranbaxy.com. Telephone (91-124), 4011832. Fax (91-124) 4011832.

- Ikawa, K.; Nomura, K.; Morikawa, N.; Ikeda, K.; Ohge, H.; Sueda, T.; Taniwaki, M. *J. Infect Chemother.* **2008**, *14*, 130–136.
- Ishikawa, T.; Iizawa, Y.; Okonogi, K.; Miyake, A. *J. Antibiot.* **2000**, *53*, 1053–1070.
- Shimizu, H.; Mikara, Y.; Doi, Y. (Takeda). U.S. Patent 4,933,334, 1990.
- Miyake, A.; Kondo, M.; Fujino, M. (Takeda). U.S. Patent 5,409,919, 1995.
- Yoshida, K.; Nishimura, S.; Maeda, S. (Takeda). JP Patent 10291993A, 1998. *Chem. Abstr.* **1998**, *130*, 24909.

## Scheme 1



**Table 1.** Preparation of cefozopran free base (**4**) under different conditions<sup>a</sup>

entry	solvent (volume with respect to input 3)	base	yield (%)
1	THF/water (5:5)	TBA	— <sup>b</sup>
2	THF/water (10:5)	TBA	91
3	THF/water (15:8)	TBA	95
4	THF/water (12:8)	TBA	90
5	THF/water 15:8	TEA	85
6	THF/water (12:8)	TMG	75

<sup>a</sup> TBA: tri-*n*-butyl amine, TMG: tetramethylguanidine, TEA: triethylamine.  
<sup>b</sup> Isolation problem.

**Preparation of (6*R*,7*R*)-7[(*Z*)-2-(5-Amino-1,2,4-thiadiazol-3-yl)-2-methoxy-iminoacetyl-amino]-3-(1*H*-imidazo[1,2-*b*]pyridazin-4-ium-1-yl-methyl)-8-oxo-5-thia-1-azabicyclo[4,2,0]-oct-2-ene-carboxylate (Cefozopran, **4**).** 7-Amino-3-[(imidazo(1,2-*b*)pyridazinium-1-yl)methyl]-3-cephem-4-carboxylatehydroiodide (**3**) (10.0 kg, 21.7 mol) was suspended in a mixture of water (80 L) and THF (150 L) at 20–25 °C. (*S*)-(2-Benzothiazolyl)-(*Z*)-2-(2-amino-1,2,4-thiadiazol-3-yl)-2-methoxyiminothio acetate (**2b**) (10.66 kg, 30.37 mol) was added at 20–25 °C followed by TBA (7.0 kg, 37.8 mol) at 20–25 °C in 20–25

min. The resulting reaction mixture was stirred for 15 h at 20–30 °C. Reaction mixture was cooled to 10–15 °C and stirred for 2 h at 10–15 °C, and the resulting white solid was filtered, washed with a mixture of THF/water (2:1) (2 × 10 L) to give **4** (10.6 kg, 95%). Chromatographic purity by HPLC 98.51%. <sup>1</sup>H NMR (DMSO-*d*<sub>6</sub>): δ 3.0–3.4 (ABq 2H, S-CH<sub>2</sub>), 3.85 (s, 3H, O-CH<sub>3</sub>), 4.98–4.99 (d, 1H, CH-6 β-lactam), 5.22–5.5 (ABq, 2H, 3-CH<sub>2</sub>), 5.6–5.65 (m, 1H, CH-7 β-lactam), 7.94–7.98 (m, 1H, azolium ring), 8.12 (br, 2H, NH<sub>2</sub>), 8.76 (s, 2H, azolium ring), 9.06 (d, 1H, azolium ring), 9.31–9.34 (d, 1H, azolium ring), 9.47–9.5 (d, 1H, CONH). MS (*m/e*): 515.

### Acknowledgment

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### Supporting Information Available

Spectral data of cefozopran free base. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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