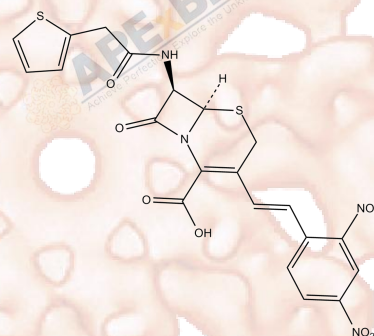


## Product Data Sheet

### Nitrocefin

<b>Cat. No.:</b>	B6052
<b>CAS No.:</b>	41906-86-9
<b>Formula:</b>	C <sub>21</sub> H <sub>16</sub> N <sub>4</sub> O <sub>8</sub> S <sub>2</sub>
<b>M.Wt:</b>	516.50
<b>Synonyms:</b>	
<b>Target:</b>	$\beta$ -lactamase
<b>Pathway:</b>	Fluorescent Dyes
<b>Storage:</b>	Store at -20° C



### Solvent & Solubility

insoluble in EtOH; insoluble in H<sub>2</sub>O;  $\geq 20.24$  mg/mL in DMSO

In Vitro

		Mass	1mg	5mg	10mg
Preparing Stock Solutions	Solvent	Concentration			
		1 mM	1.9361 mL	9.6805 mL	19.3611 mL
		5 mM	0.3872 mL	1.9361 mL	3.8722 mL
		10 mM	0.1936 mL	0.9681 mL	1.9361 mL

Please refer to the solubility information to select the appropriate solvent.

### Biological Activity

Shortsummary

Nitrocefin (CAS 41906-86-9) is a chromogenic cephalosporin substrate commonly employed in biochemical assays to detect  $\beta$ -lactamase enzymatic activity.  $\beta$ -lactamases, enzymes prevalent in microbial species, catalyze the hydrolysis of  $\beta$ -lactam antibiotics such as penicillins and cephalosporins, causing antibiotic resistance. Upon cleavage by  $\beta$ -lactamase enzymes, nitrocefin undergoes a measurable colorimetric shift from yellow to red, enabling visual or spectrophotometric detection in the range of approximately 380 – 500 nm. Nitrocefin prominently serves in microbiological and clinical research to evaluate resistance profiles of bacterial isolates and in assays for inhibitor activity toward  $\beta$ -lactamases. Its reported IC<sub>50</sub> varies depending on  $\beta$ -lactamase type, concentration, and assay conditions, with typical values ranging from approximately 0.5 to 25  $\mu$ M.

IC <sub>50</sub> & Target	
In Vitro	<b>Cell Viability Assay</b>
	Cell Line: E. coli
	Preparation method: Isogenic E. coli strains were grown to log phase at an OD600 of ~0.5 in CAMHB supplemented with 10 µg/mL chloramphenicol. Cells from 1 mL culture were harvested by centrifugation. The cell pellet was freeze-thawed, resuspended in 100 µL BugBuster HT Protein Extraction Reagent, and incubated at room temperature for 15 min with shaking, followed by centrifugation at 21,100 × g for 20 min. The supernatant (cell lysate) was taken and used in the Bradford assay to measure the protein concentration and in the β-lactamase activity assay. In the β-lactamase activity assay (total reaction volume: 150 µL), cell lysate (5 µL) was incubated with 100 µM nitrocefin in 1 × phosphate-buffered saline (pH 7.4) containing 0.1 mg/mL bovine serum albumin (BSA), and absorbance at 486 nm was measured for 30 min. β-Lactamase-specific activity (nanomoles of nitrocefin hydrolyzed per minute per milligram of protein) was determined using the nitrocefin extinction coefficient of 17,400 M <sup>-1</sup> cm <sup>-1</sup>
	Reacting conditions: 5 µL cell lysate (E. coli at an OD600 of ~0.5), 100 µM nitrocefin in 1 × phosphate-buffered saline (pH 7.4, 0.1 mg/mL bovine serum albumin (BSA), measured for 30 min at 486 nm
In Vivo	Applications: The β-lactamase activity of the lysate from E. coli cells overproducing wild-type PenI was 600-fold more active than that from cells overproducing the C69Y variant, consistent with the biochemical data using the purified proteins.
	<b>Animal experiment</b>
	Applications:
In Vivo	Other notes: The technical data provided above is for reference only. The reagent is basically applied in Nitrocefin β-lactamase assay for bacteria.

## Product Citations

See more customer validations on [www.apexbt.com](http://www.apexbt.com).

## References

1.Mojica MF, Becka SA, Edwards M, Myers C, Uehara K, Uehara T, Hoshino T, Zeiser ET, Chatwin CL, Six DA, Bonomo RA, Papp-Wallace KM, Nukaga M. O. Burkholderia pseudomallei PenI β-lactamase and variants are potently inhibited by taniborbactam. Antimicrob Agents Chemother 0:e00787-25.

## Caution

**FOR RESEARCH PURPOSES ONLY.**

**NOT FOR HUMAN, VETERINARY DIAGNOSTIC OR THERAPEUTIC USE.**

*Specific storage and handling information for each product is indicated on the product datasheet. Most APExBIO products are stable under the recommended conditions. Products are sometimes shipped at a temperature that differs from the recommended storage temperature. Shortterm storage of many products are stable in the short-term at temperatures that differ from that required for long-term storage. We ensure that the product is shipped under conditions that will maintain the quality of the reagents. Upon receipt of the product, follow the storage recommendations on the product data sheet.*

**APExBIO Technology**

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