

# Human CXCL8 knockout PC-3 cell line ab273743

5 Images

## Overview

<b>Product name</b>	Human CXCL8 knockout PC-3 cell line
<b>Parental Cell Line</b>	PC3
<b>Organism</b>	Human
<b>Mutation description</b>	Knockout achieved by using CRISPR/Cas9, Homozygous: 22% 11 bp deletion, 24% 7 bp deletion, 54% 2 bp deletion in exon 2
<b>Passage number</b>	<20
<b>Knockout validation</b>	Next Generation Sequencing (NGS), Western Blot (WB)
<b>Tested applications</b>	<b>Suitable for:</b> WB, Sandwich ELISA
<b>Biosafety level</b>	1
<b>General notes</b>	<p><b>Recommended control:</b> Human wild-type PC3 cell line (<a href="#">ab275472</a>). Please note a wild-type cell line is not automatically included with a knockout cell line order, if required please add recommended wild-type cell line at no additional cost using the code WILDTYPE-TMTK1.</p> <p><b>Cryopreservation cell medium:</b> Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose.</p> <p><b>Culture medium:</b> F-12K + 10% FBS</p> <p><b>Initial handling guidelines:</b> Upon arrival, the vial should be stored in liquid nitrogen vapor phase and not at -80°C. Storage at -80°C may result in loss of viability.</p> <ol style="list-style-type: none"> <li>1. Thaw the vial in 37°C water bath for approximately 1-2 minutes.</li> <li>2. Transfer the cell suspension (0.8 mL) to a 15 mL/50 mL conical sterile polypropylene centrifuge tube containing 8.4 mL pre-warmed culture medium, wash vial with an additional 0.8 mL culture medium (total volume 10 mL) to collect remaining cells, and centrifuge at 201 x g (rcf) for 5 minutes at room temperature. 10 mL represents minimum recommended dilution. 20 mL represents maximum recommended dilution.</li> <li>3. Resuspend the cell pellet in 5 mL pre-warmed culture medium and count using a haemocytometer or alternative cell counting method. Based on cell count, seed cells in an appropriate cell culture flask at a density of <math>2 \times 10^4</math> cells/cm<sup>2</sup>. Seeding density is given as a guide only and should be scaled to align with individual lab schedules.</li> <li>4. Incubate the culture at 37°C incubator with 5% CO<sub>2</sub>. Cultures should be monitored daily.</li> </ol> <p><b>Subculture guidelines:</b></p> <p>All seeding densities should be based on cell counts gained by established methods. A guide seeding density of <math>2 \times 10^4</math> cells/cm<sup>2</sup> is recommended.</p> <p>A partial media change 24 hours prior to subculture may be helpful to encourage growth, if</p>

required.

Cells should be passaged when they have achieved 80-90% confluence.

This product is subject to limited use licenses from The Broad Institute and ERS Genomics Limited, and is developed with patented technology. For full details of the limited use licenses and relevant patents please refer to our [limited use license](#) and [patent pages](#).

## Properties

---

<b>Number of cells</b>	1 x 10 <sup>6</sup> cells/vial, 1 mL
<b>Viability</b>	~80%
<b>Adherent /Suspension</b>	Adherent
<b>Tissue</b>	Prostate
<b>Cell type</b>	epithelial
<b>Disease</b>	Adenocarcinoma
<b>Gender</b>	Male
<b>Mycoplasma free</b>	Yes
<b>Storage instructions</b>	Shipped on Dry Ice. Store in liquid nitrogen.
<b>Storage buffer</b>	Constituents: 8.7% Dimethylsulfoxide, 2% Cellulose, methyl ether

## Target

---

<b>Function</b>	IL-8 is a chemotactic factor that attracts neutrophils, basophils, and T-cells, but not monocytes. It is also involved in neutrophil activation. It is released from several cell types in response to an inflammatory stimulus. IL-8(6-77) has a 5-10-fold higher activity on neutrophil activation, IL-8(5-77) has increased activity on neutrophil activation and IL-8(7-77) has a higher affinity to receptors CXCR1 and CXCR2 as compared to IL-8(1-77), respectively.
<b>Sequence similarities</b>	Belongs to the intercrine alpha (chemokine CxC) family.
<b>Post-translational modifications</b>	Several N-terminal processed forms are produced by proteolytic cleavage after secretion from at least peripheral blood monocytes, leukocytes and endothelial cells. In general, IL-8(1-77) is referred to as interleukin-8. IL-8(6-77) is the most prominent form.
<b>Cellular localization</b>	Secreted.

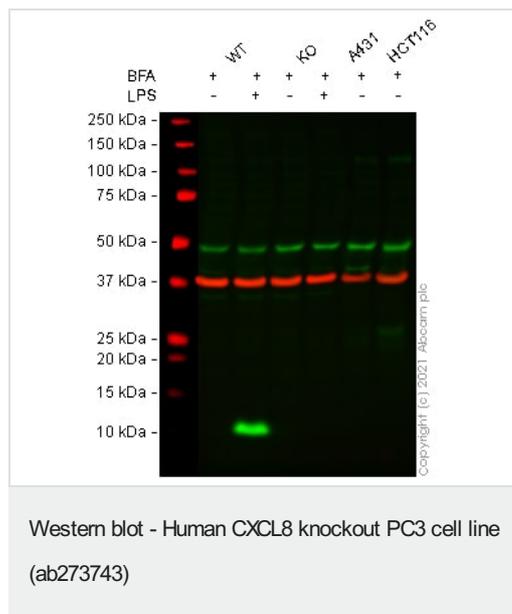
---

## Applications

---

**The Abpromise guarantee** Our [Abpromise guarantee](#) covers the use of ab273743 in the following tested applications. The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		Use at an assay dependent concentration.
Sandwich ELISA		Use at an assay dependent concentration.



**All lanes** : Anti-IL-8 antibody [EPR22994-255] (**ab235584**) at 1/1000 dilution

**Lane 1** : Wild-type PC-3 Brefeldin A (**ab120299**)-treated (5 µg/ml, 5 h) cell lysate

**Lane 2** : Wild-type PC-3 LPS-treated (2 µg/ml, 6 h) with Brefeldin A (**ab120299**) (5 µg/ml, 5 h) cell lysate

**Lane 3** : CXCL8 knockout PC-3 Brefeldin A (**ab120299**)-treated (5 µg/ml, 5 h) cell lysate

**Lane 4** : CXCL8 knockout PC-3 LPS-treated (2 µg/ml, 6 h) with Brefeldin A (**ab120299**) (5 µg/ml, 5 h) cell lysate

**Lane 5** : A431 cell lysate

**Lane 6** : HCT116 cell lysate

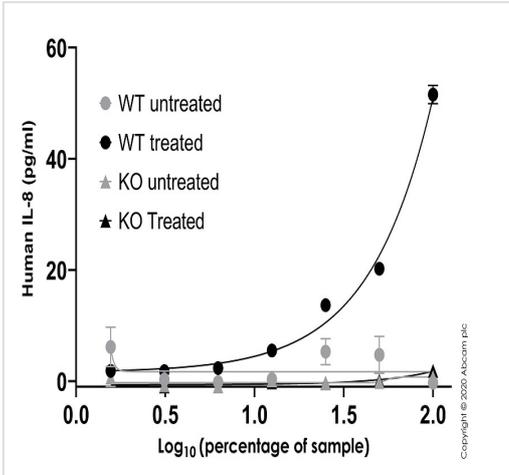
Lysates/proteins at 30 µg per lane.

Performed under reducing conditions.

**Observed band size:** 10 kDa

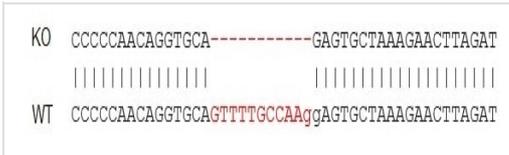
**Lanes 1 - 6:** Merged signal (red and green). Green - **ab235584** observed at 10 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

**ab235584** was shown to react with IL-8 in wild-type PC-3 cells in Western blot with loss of signal observed in CXCL8 knockout cell line ab273743 (knockout cell lysate **ab275520**). Wild-type PC-3 and CXCL8 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab235584** and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



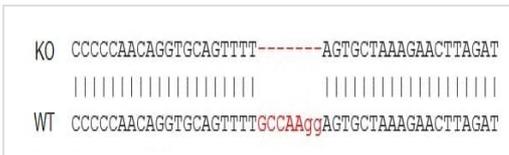
Sandwich ELISA - Human CXCL8 knockout PC-3 cell line (ab273743)

Human IL-8 concentration was interpolated from the standard curve. Supernatants from cell culture samples were serially diluted and assessed by the Human IL-8 ELISA kit ([ab214030](#)). Wild-type PC-3 cells and CXCL8 knockout PC-3 cells (ab273743) were assessed in duplicate (n=2) and were either treated with 2 µg/ml LPS for 6 hours to induce expression of IL-8 or not treated with LPS. Data are represented as the mean and error bars represent standard deviation.



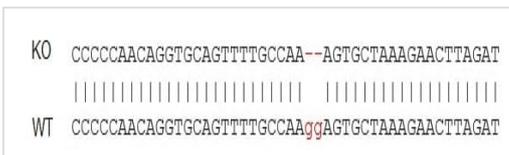
Next Generation Sequencing - Human CXCL8 (IL-8) knockout PC3 cell line (ab273743)

Allele-1: 11bp deletion in exon 2.



Next Generation Sequencing - Human CXCL8 (IL-8) knockout PC3 cell line (ab273743)

Allele-2: 7bp deletion in exon 2.



Next Generation Sequencing - Human CXCL8 (IL-8) knockout PC3 cell line (ab273743)

Allele-3: 2bp deletion in exon 2.

**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

## **Our Abpromise to you: Quality guaranteed and expert technical support**

---

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
  
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit <https://www.abcam.com/abpromise> or contact our technical team.

## **Terms and conditions**

---

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors