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Data Sheet

CD22 / Luciferase - CHO Recombinant Cell Line
Catalog #79715

Description
Recombinant clonal stable CHO cell line constitutively expressing full length human CD22 protein (Genbank #NM_001771) and the firefly luciferase. Surface expression of CD22 was confirmed by flow cytometry.

Background
CD22, also known as Siglec-2, is expressed on the membrane of B-cells. It is reported to act as an inhibitory co-receptor of the B-cell receptor to control the body's B-cell response. In 2017 the FDA approved inotuzumab ozogamicin (Besponsa), an antibody-drug conjugate targeting CD22, for patients with B-cell acute lymphoblastic leukemia (ALL). Additional therapies targeting CD22 are under evaluation.

Application
Useful for screening and validating antibodies against CD22 and different CD22 CAR-T for immunotherapy research and drug discovery. Also useful for CD22 binding assays to screen for CD22 ligands.

Host Cell
CHO K1 cell line, Chinese Hamster Ovary

Format
Each vial contains ~2 x 10^6 cells in 1 ml of 10% DMSO in FBS.

Storage
Store in liquid nitrogen immediately upon receipt.

Materials Required but Not Supplied
• Thaw Medium 3 (BPS Bioscience, #60186)
• Growth Medium 3A (BPS Bioscience, #60188)
• 96-well tissue culture-treated white clear-bottom assay plate
• ONE-Step luciferase assay system (BPS Bioscience, #60690) or other luciferase reagent for measuring firefly luciferase activity
• Luminometer

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Cell Culture
Thaw Medium 3 (BPS Bioscience, #60186): F-12K Medium supplemented with 10% FBS, 1% Penicillin/Streptomycin

Growth Medium 3A (BPS Bioscience, #60188): F-12K Medium supplemented with 10% FBS, 1% Penicillin/Streptomycin plus 1 mg/ml G418 and 500 µg/ml of Hygromycin B to ensure recombinant expression.

Cells should be grown at 37˚C with 5% CO_2 using Growth Medium 3A. CD22/Luciferase CHO cells should exhibit a typical cell division time of ~24 hours**.**

It is recommended to quickly thaw the frozen cells from liquid nitrogen in a 37°C water bath, transfer to a tube containing 10 ml of **Thaw Medium 3 (no Geneticin or Hygromycin B),** spin the cells down, remove the supernatant, and then re-suspend the cells in pre-warmed Thaw Medium 3 **(no Geneticin or Hygromycin B).** Then transfer the re-suspended cells to a T25 flask and culture in a 37˚C CO_2 incubator overnight. The next day, replace the medium with fresh Thaw Medium 3 **(no Geneticin or Hygromycin B)** and continue growing in a CO_2 incubator at 37°C until the cells are ready to be split. Cells should be split before they reach complete confluence. After the first passage, switch to **Growth Medium 3A** (contains **Geneticin and Hygromycin B**).

To passage the cells, rinse the cells with Phosphate Buffered Saline (PBS), detach the cells from the culture vessel with 0.25% Trypsin/EDTA, and add Growth Medium 3A and transfer to a tube. Next, spin the cells down, remove the supernatant, and then re-suspend the cells and seed appropriate aliquots of the cell suspension into new culture vessels. Suggested subcultivation ratios: 1:10 to 1:20 twice a week.

To freeze the cells down, rinse the cells with Phosphate Buffered Saline (PBS), and detach the cells from the culture vessel with 0.25% Trypsin/EDTA. After detachment, add **Thaw Medium 3 (no Geneticin or Hygromycin B)** and count the cells, then transfer to a tube, spin the cells down, and resuspend in 4˚C Freezing Medium (10% DMSO + 90% FBS) at ~2 x 10^6 cells/ml. Dispense 1 ml of cell aliquots into each cryogenic vial. Place vials in an insulated container for slow cooling and store at -80˚C overnight. Transfer to liquid nitrogen the next day for storage. It is recommended to expand the cells and freeze down more than 10 vials of cells for future use at early passages.

Mycoplasma Testing
This cell line has been screened using the MycoAlert™ Mycoplasma Detection Kit (Lonza, #LT07-118) to confirm the absence of Mycoplasma contamination. MycoAlert Assay Control Set (Lonza, #LT07-518) was used as a positive control.

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Application References

1. Poe J, *et al.* CD22 and Siglec-G in B cell function and tolerance. *Trends Immunol. 2012* Aug; **33(8):**413-420.
2. Dorner T, *et al.* The mechanistic impact of CD22 engagement with epratuzumab on B cell function: Implications for the treatment of systemic lupus erythematosus. *Autoimmunity Reviews.* 2015 **14:**1079-1086.
3. Wei G., *et al.* Novel immunotherapies for adult patients with B-lineage acute lymphoblastic leukemia. *Journal of Hematology and Oncology.* 2017 **10:**1-13.

Vector and Sequence

Human CD22 (NM_001771) was cloned into pIRESneo3.

TYRALDGDLESFILFHNPEYNKNTSKFDGTRLYESTKDGKVPSEQKRVQFLGDKNKNCTLSIHP
VHLNDSGQLGLRMESKTEKWMERIHLNVSERPFPPHIQLPPEIQESQEVTLTCLLNFSCYGYPI
QLQWLLEGVPMRQAAVTSTSLTIKSVFTRSELKFSPQWSHHGKIVTCQLQDADGKFLSNDTVQ
LNVKHTPKLEIKVTPSDAIVREGDSVTMTCEVSSSNPEYTTVSWLKDGTSLKKQNTFTLNLREV
TKDQSGKYCCQVSNDVGPGRSEEVFLQVQYAPEPSTVQILHSPAVEGSQVEFLCMSLANPLP
TNYTWYHNGKEMQGRTEEKVHIPKILPWHAGTYSCVAENILGTGQRGPGAELDVQYPPKKVTT
VIQNPMPIREGDTVTLSCNYNSSNPSVTRYEWKPHGAWEEPSLGVLKIQNVGWDNTTIACAAC
NSWCSWASPVALNVQYAPRDVRVRKIKPLSEIHSGNSVSLQCDFSSSHPKEVQFFWEKNGRL
LGKESQLNFDSISPEDAGSYSCWVNNSIGQTASKAWTLEVLYAPRRLRVSMSPGDQVMEGKS
ATLTCESDANPPVSHYTWFDWNNQSLPYHSQKLRLEPVKVQHSGAYWCQGTNSVGKGRSPL
STLTVYYSPETIGRRVAVGLGSCLAILILAICGLKLQRRWKRTQSQQGLQENSSGQSFFVRNKK
VRRAPLSEGPHSLGCYNPMMEDGISYTTLRFPEMNIPRTGDAESSEMQRPPPDCDDTVTYSA
LHKRQVGDYENVIPDFPEDEGIHYSELIQFGVGERPQAQENVDYVILKH

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Quality Assurance

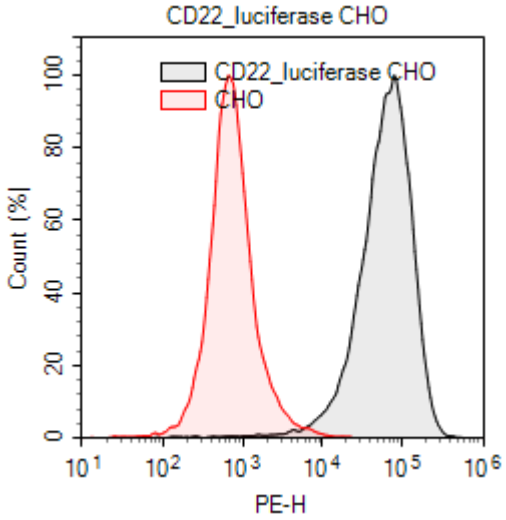

Figure 1. Expression of CD22 validated by flow cytometry. Flow cytometry using PE-conjugated anti-human CD22 antibody (Biolegend, #302506) to detect CD22 surface expression on either CD22/Luciferase CHO cells (black) or parental CHO-K1 cells (red).

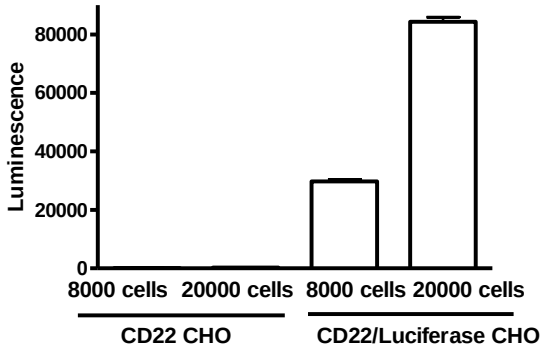

Figure 2. Luciferase activity of CD22/Luciferase CHO recombinant cells.
CD22/Luciferase CHO recombinant cells were seeded in a 96-well plate at 8000 cells/well or 20000 cells/well. The next day, luciferase activity was measured using the ONE-Step luciferase assay system (BPS Bioscience, #60690).

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Related Products

Product	Cat. #	Size
Thaw Medium 3	60186	100 ml
Growth Medium 3A	60188	500 ml
CD22, Fc-fusion (IgG1), Avi-Tag HiP™	79464	100 µg
CD22, Fc-fusion (IgG1), Avi-Tag HiP™	79466	50 µg
CD22 CHO Recombinant Cell Line (Medium Expression)	79557-M	2 vials
CD22 CHO Recombinant Cell Line (High Expression)	79557-H	2 vials
BCMA/Luciferase-CHO Recombinant Cell line	79724	2 vials
CD19/Luciferase-CHO Recombinant Cell Line	79714	2 vials
BCMA— CHO Recombinant Cell Line (High Expression)	79500-H	2 vials
BCMA— CHO Recombinant Cell Line (Medium Expression)	79500-M	2 vials
BCMA— CHO Recombinant Cell Line (Low Expression)	79500-L	2 vials