abcam

Product datasheet

Human AGO2 knockout MCF7 cell line ab287203

Overview

Product name	Human AGO2 knockout MCF7 cell line
Parental Cell Line	MCF7
Organism	Human
Passage number	<20
Biosafety level	1
General notes	Although we aim to provide customers with a homozygous clone, feasibility will be dependent on the biology of the protein. Should only heterozygous edits be achieved, you will be notified of the outcome and be asked to confirm whether the cell line is acceptable. All clones will be accompanied with DNA sequencing data, and the mutation description. **Recommended control**: Human wild-type MCF7 cell line (**ab288560**). 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. **Cryopreservation cell medium**: Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose. **Culture medium**: MEM + 10% FBS + 0.01 mg/mL bovine insulin **Initial handling guidelines**: 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. 1. Thaw the vial in 37°C water bath for approximately 1-2 minutes. 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. 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 5-7x10^4 cells/cm2. Seeding density is given as a guide only and should be scaled to align with individual lab schedules. 4. Incubate the culture at 37°C incubator with 5% CO2. Cultures should be monitored daily. **Subculture guidelines**: All seeding densities should be based on cell counts gained by established methods. A guide seeding density of 5-7x10^4 cells/cm2 is recommended. A partial media change 24 hours prior to subculture may be helpful to encourage growth, if required. Cells should be passaged when they have achieved 80-90% confluence.

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We will provide viable cells that proliferate on revival.

Properties

Number of cells	1000000 cells/vial, 1 mL
Adherent /Suspension	Adherent
Tissue	Breast
Cell type	epithelial
Disease	Adenocarcinoma
Gender	Female
Mycoplasma free	Yes
Storage instructions	Shipped on Dry Ice. Store in liquid nitrogen.
Storage buffer	Constituents: 8.7% Dimethylsulfoxide, 2% Cellulose, methyl ether

Target

Function	Required for RNA-mediated gene silencing (RNAi) by the RNA-induced silencing complex (RISC). The 'minimal RISC' appears to include EIF2C2/AGO2 bound to a short guide RNA such as a microRNA (miRNA) or short interfering RNA (siRNA). These guide RNAs direct RISC to complementary mRNAs that are targets for RISC-mediated gene silencing. The precise mechanism of gene silencing depends on the degree of complementarity between the miRNA or siRNA and its target. Binding of RISC to a perfectly complementary mRNA generally results in silencing due to endonucleolytic cleavage of the mRNA specifically by EIF2C2/AGO2. Binding of RISC to a partially complementary mRNA results in silencing through inhibition of translation, and this is independent of endonuclease activity. May inhibit translation initiation by binding to the 7-methylguanosine cap, thereby preventing the recruitment of the translation initiation factor eIF4-E. May also inhibit translation initiation via interaction with EIF6, which itself binds to the 60S ribosomal subunit and prevents its association with the 40S ribosomal subunit. The inhibition of translational initiation leads to the accumulation of the affected mRNA in cytoplasmic processing bodies (P-bodies), where mRNA degradation may subsequently occur. In some cases RISC-mediated translational repression is also observed for miRNAs that perfectly match the 3' untranslated region (3'-UTR). Can also upregulate the translation of specific mRNAs under certain growth conditions. Binds to the AU element of the 3'-UTR of the TNF (TNF-alpha) mRNA and upregulates translation under conditions of serum starvation. Also required for transcriptional gene silencing (TGS), in which short RNAs known as antigene RNAs or agRNAs direct the transcriptional repression of complementary promoter regions.
Sequence similarities	Belongs to the argonaute family. Ago subfamily. Contains 1 PAZ domain. Contains 1 Piwi domain.
Domain	The Piwi domain may perform RNA cleavage by a mechanism similar to that of RNase H. However while RNase H utilizes a triad of Asp-Asp-Glu (DDE) for metal ion coordination, this

	protein appears to utilize a triad of Asp-Asp-His (DDH).
Post-translational modifications	Hydroxylated. 4-hydroxylation appears to enhance protein stability but is not required for miRNA-binding or endonuclease activity.
Cellular localization	Cytoplasm > P-body. Nucleus. Translational repression of mRNAs results in their recruitment to P-bodies. Translocation to the nucleus requires IMP8.

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