

## 14-510ACL: Cas9 Stable Cell Line

**Application :** Functional Assay

### Description

Cas9 Stable Cell Line is a stably transfected HEK 293 cell line which expresses CRISPR associated protein 9 (Cas9, also known as Csn1), an RNA-guided DNA endonuclease enzyme associated with the Clustered Regularly Interspersed Short Palindromic Repeats (CRISPR) from *Streptococcus pyogenes*.

**Sequence data:** Cas9 (accession number WP\_010922251)

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MDKKYSIGLDIGTNSVGWAVITDEYKVPSSKFKVLGNTDRHSIKKNLIGALLFDSGETAE
ATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFHRLLEESFLVEEDKKHERHPIFG
NIVDEVAYHEKYPTIYHLRKKLVDSTDKADLRLLIYLALAHMIKFRGHFLIEGDLNPDNSD
VDKLFQLVQTYNQLFEENPINASGVDAKAILSARLSKSRLENLIAQLPGEKKNGLFGN
LIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLNLLAQIGDQYADLFLAAKNLSDAI
LLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKYKEIFFDQSKNGYA
GYIDGGASQEEFYKFIKPILEKMDGTEELLVKNREDLLRKQRTFDNGSIPHQIHLGELH
AILRRQEDFYFPLKDNREKIEKILTRIPYYVGPLARGNSRFAWMTRKSEETITPWNFEE
VVDKGASAQSFIERMTNFDKNLPNEKVLPHKSLLEYFTVYNELTKVKYVTEGMRKPAFL
SGEQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTYHDLLKI
IKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKYAHLFDDKVMKQLKRRRYTGWG
RLSRKLINGIRDKQSGKTILDFLKSDFANRNFMLIHDDSLTFKEDIQKAQVSGQGDSL
HEHIANLAGSPAIKKILQTVKVVDELVKVMGRHKPENIVIAMARENQTTQKGQKNSRER
MKRIEEGKELGSQILKEHPVENTQLQNEKLYLYLQNGRDMYVDQELDINRLSDYDVDH
IVPQSFLKDDSIDNKVLRSDKNRKGSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNL
TKAERGGLSELDKAGFIKRLVETROITKHVAQILDSRMNTKYDENDKLIREVKVITLKS
KLVSDFRKDFQFYKREINNYHHAHDAYLNAVVGTAIIKYPKLESEFVYGDYKVYDVRK
MIAKSEQEIGKATAKYFFYSNIMNFFKTEITLANGEIRKRPLIETNGETGEIVWDKGRDF
ATVRKVLSPQVNVKTEVQTGGFSKESILPKRNSDKLIARKKDWDPKKYGGFDSPTVA
YSVLVAKVEKGSKKLKSVKELLGITIMERSSEFEKNPIDFLEAKGYKEVKKDLIILPK
YSLFELENGRKRMLASAGELQKGNELALPSKYVNFYLAHYEKLKGSPEDEQKQLFVE
QHKHYLDEIIEQISEFSKRVLADANLDKVL SAYNKHRDKPIREQAENIIHLFTLTNLGA
PAAFKYFDTTIDRKRYTSTKEVLDTLIHQISITGLYETRIDLSQLGGD
```

### Product Info

**Amount :** 1 Vial  
**Content :** Each vial contains  $2 \sim 3 \times 10^6$  cells in 1 ml of 90% FBS + 10% DMSO  
**Storage condition :** Immediately upon receipt, store in liquid nitrogen.

### Application Note

**Application:**

- Screen for antibodies of Cas9 through Western blotting.

### Culture conditions:

Cells should be grown at 37°C with 5% CO<sub>2</sub> using DMEM medium (w/ L-Glutamine, 4.5g/L Glucose and Sodium Pyruvate) supplemented with 10% heat-inactivated FBS and 1% Pen/Strep, plus 10 µg/ml of Blastcidin.

It is recommended to quickly thaw the frozen cells upon receipt or from liquid nitrogen in a 37°C water-bath, transfer to a tube containing 10 ml of growth medium without Blastcidin, spin down cells, resuspend cells in pre-warmed growth medium without Blastcidin, transfer resuspended cells to T25 flask and culture in 37°C-CO<sub>2</sub> incubator.

Leave the T25 flask in the incubator for 1~3 days without disturbing or changing the medium until cells completely recover viability and become adherent. Once cells are over 90% adherent, remove growth medium and passage the cells through trypsinization and centrifugation. At first passage, switch to growth medium containing Blastcidin. Cells should be split before they reach complete confluence.

To passage the cells, detach cells from culture vessel with Trypsin/EDTA, add complete growth medium and transfer to a tube, spin down cells, resuspend cells and seed appropriate aliquots of cells suspension into new culture vessels. Subcultivation ration = 1:10 to 1:20 weekly. To achieve satisfactory results, cells should not be passaged over 16 times.

### LIMITED USE RESTRICTIONS:

**THIS PRODUCT IS SOLELY FOR IN VITRO RESEARCH USE ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.**

**By use of this product, user agrees to be bound by the terms of this limited use statement.**

**This product is solely for Internal Research Purposes and not for Commercial Purposes. Commercial Purposes include, but are not limited to (1) use of the cell line in manufacturing; (2) use of the cell line to provide a service, information or data; (3) use of the cell line for therapeutic, diagnostic or prophylactic purposes; or (4) resale of the cell line whether or not such cell lines are resold for use in research. The buyer cannot sell, give or otherwise transfer this product to a third party.**

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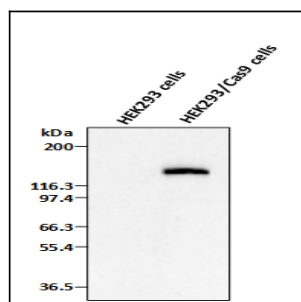


Fig-1: Detection of Cas9 in the HEK293/Cas9 stable cell line by Western blotting using anti-Cas9 antibody.