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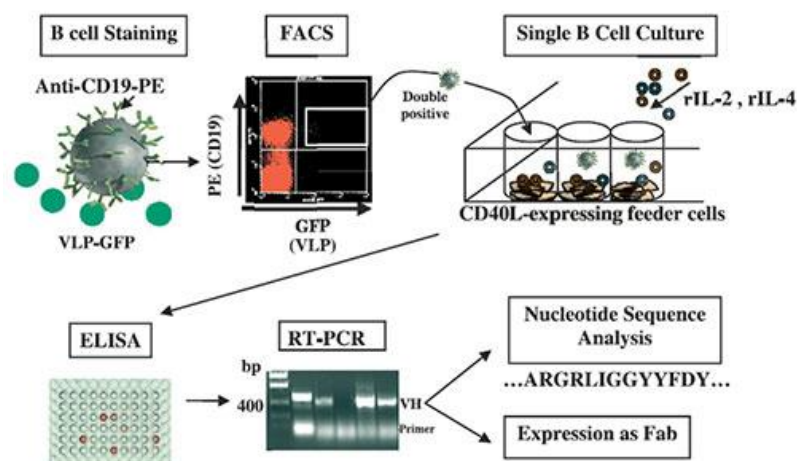
# Product Manual

## CD40L Expressing Feeder Cell Line (EL-4-5B)

Catalog #	Size	Product Type
C1001	1 vial	Cell line

**Alternative name** EL-4-5B

**Description** The EL-4-B5 cells express CD40-ligand. EL4-B5 is grown with B cells to activate the B cells via direct cell contact to induce proliferation, differentiation, and secretion of antibody. The EL-4-B5 cell line offers a method for the generation of recombinant human mAbs from single antigen-specific B cell clones selected with fluorescent VLPs and can be used to generate human mAbs to many other viruses whose proteins can self-assemble into VLPs.



**Adapted from:** Weitkamp J-H., et al. J Immunol Methods 2003, 275:223-237.

**Cell Type** Mouse thymoma continuous cell line

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<b>Accession ID</b>	CVCL_5139
<b>Organism</b>	Mouse
<b>Biosafety Level</b>	2
<b>Subculturing</b>	Cells are both adherent and suspension. The suspension cells are the more healthy ones. Can tolerate large splits after line is established (volume ratios of 1:10, for example). Helper function is optimal when cells are in log phase growth. Keep density below $10^6$ cells per mL at all times
<b>Maintenance Tips</b>	<ul style="list-style-type: none"><li>✓ It is not unusual at all for the vibrant culture counted just after thawing to take a dismal turn within a short 24-hours to what appears to be complete failure. Generally speaking, these cells may take a week or so, but the culture rebounds and becomes very difficult to keep up with because it grows so robustly. If in doubt, scrape the walls of the flask to remove the adherent phenotype; Stand the culture flask up in the incubator for ~1 hour, to allow cells to settle to the bottom; remove an arbitrary volume of medium from the 'top' of the culture, so that the total volume of medium is obviously reduced, forcing the cells closer together and hopefully stimulating the tactile contact that budding cultures need.</li><li>✓ Most lymphocytes in culture - in the same culture flask, exhibit two phenotypes: they float about in little Volvox-like balls or they pile on top of each other, with the cells on the bottom of the pile sticking to the plastic of the flask. The charge of the plastic is the accounts for this. There is no need to trypsinize the adherent cells, but they can be scraped gently with a cell scraper, if you want to bother with them at all. Once my cultures were established, the cells were generally had from the suspended ones. When using the cells or just passaging them, you can ignore the adherent ones.</li></ul>

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- ✓ B cell helper function depends on the batch of FBS used. There is no simple way to identify optimal FBS lots, other than trial and error with the final assay (typically human B cell expansion).

## Freezing Protocol

1. Count log phase cells.
2. Centrifuge cell suspension @1700rpm, 5 min, RT.
3. Gently resuspend cell pellet in freezing medium at a density of  $2 \times 10^6$ /mL.
4. Distribute one microliter aliquots into 2mL cryovials.
5. Place filled cryovials into freezer container and place -70°C overnight.
6. Next day, the cryovials may be moved to vapor phase of a liquid nitrogen freezer for long term storage.
7. Alternatively, the cryovials can be put into a liquid nitrogen freezer directly at vapor phase, and moved into liquid phase after 24hrs.

**Freezing Medium:** 90%-FCS, 10% DMSO

**Make fresh for each use.**

## Note

For laboratory research only. Not for clinical applications.

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