### Designation:
**P-815**

### CLS order number:
- Cryovial: 400242
- Vital: 440242

### Origin and General Characteristics
- **Depositor:** Ralph
- **Organism:** *Mus musculus* (mouse)
- **Strain:** DBA/2
- **Tissue:** Mastocytoma
- **Morphology:** Round cells
- **Cell type:** Mast cell
- **Growth Properties:** Suspension (some adherent cells)
- **Description:** P-815 cells phagocytose latex beads but not zymosan or BCG. They do not function in antibody dependent cell mediated cytotoxicity. Growth of the cells is not inhibited by dextran sulfate, LPS or PPD. Tested and found negative for ectromelia virus (mousepox).

### Culture Conditions and Handling
- **Culture Medium:** DMEM supplemented with 4.5 g/L glucose, 2 mM L-glutamine, and 10% fetal bovine serum (MG-30, CLS order number 820300).
- **Subculturing:** Start cultures at 2x10^5 cells/ml and maintain between 1x10^5 and 1x10^6 cells/ml. Subculture by transferring an appropriate aliquot of the suspension into new flasks filled with fresh cell culture medium. Detach adherent cells by washing with PBS first and incubation with Accutase at ambient temeratur 8-10 minutes. Combine with the non-adherent cells and distribute into new cell culture flasks.
- **Fluid Renewal:** Every 2 to 3 days
- **Freeze Medium:** CM-2 (CLS order number: 800225, 25ml, 800250, 50ml)
- **Sterility:** Mycoplasma specific PCR: negative; Bacteria specific PCR: negative
- **Biosafety Level:** 1

### Special Features of the Cell Line
- **Products:** Lysozyme

### References:
Recommendations for handling of adherent cell cultures following delivery

Cryopreserved cells
If immediate culturing is not intended, the cryovial(s) must be stored in liquid nitrogen after arrival.
If immediate culturing is intended, please follow these instructions:
Quickly thaw by rapid agitation in a 37°C water bath within 40-60 seconds. The water bath should have clean water containing an antimicrobial agent. As soon as the sample has thawed, remove the cryovial from the water bath (a small ice clump should remain and the cryovial should still be cold).
From now on, all operations should be carried out under aseptic conditions.
Immediately transfer the cryovial to a sterile flow cabinet and wipe with 70% alcohol. Carefully open the vial and transfer the cell suspension into a 15 ml centrifuge tube containing 8 ml of culture medium (room temperature, not 37°C). Resuspend the cells carefully. The cells may be spun down at 300xg for 3 minutes. After centrifuging, aseptically remove the supernatant and add 10 ml of fresh cell culture media. Carefully resuspend the cells and distribute into two 25cm² cell culture flask. Incubate at 37°C/5% CO₂.
Subculture as soon as the cell concentration has reached 1 x 10⁶ cells/ml or the cells reach a 90% confluency. It is recommended to distribute the cells into new flasks containing fresh medium thus diminishing the amount of dead cells and cell debris. Adjust to a cell concentration of 1-2 x 10⁵ cells/ml depending on the specification given for the cell line.
After about 1-2 times of sub-culturing as recommended the percentage of viable cells should be > 90%.

Proliferating Cultures
Immediately after receipt, the cell concentration should be determined. If the cell concentration already has reached a value of 1 x 10⁶ cells/ml or even more, subculture the cells as described above. Remove the entire content of the flask and centrifuge at 300xg for 3 minutes.
Resuspend the cell pellets as suggested under subculture procedures described on the appropriate datasheet.

Safety precautions for frozen cell lines
If the cryovial is planned to be stored in liquid nitrogen and to be thawed in the future, special safety precautions should be followed:
- Protective gloves and clothing should be used and a facemask or safety goggles must be worn when storing and/or thawing the cryovial.
- The removal of a cryovial from liquid nitrogen can result in the explosion of the cryovial creating flying fragments.