

## Data Sheet

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<b>BCRJ Code:</b>	0404
<b>Cell Line:</b>	Kyse-30
<b>Species:</b>	Homo sapiens
<b>Vulgar Name:</b>	Human
<b>Tissue:</b>	Esophagus
<b>Cell Type:</b>	Polygonal
<b>Morphology:</b>	Epitheloid with long processes growing in monolayers
<b>Disease:</b>	Squamous Carcinoma
<b>Growth Properties:</b>	Adherent
<b>Sex:</b>	Male
<b>Age/Ethnicity:</b>	64 Year / Asian
<b>Derivation:</b>	KYSE-30 was established from the oesophageal cancer of an untreated 64 year old male. The tumour sample was taken from the mucosal surface of a well differentiated squamous cell carcinoma. The cell line KYSE-30 was established with the use of tumours initially transplanted to athymic mice. The cells are reported to have a doubling time of 20.8 hrs in the exponential growth phase. A p53 mutation at the splice acceptor site of intron 6 and a 12 fold amplification of c-erb B has been reported. KYSE-30 cells express a large number of epidermal growth factor receptors, 1.2x10,000,000 sites/cell.
<b>Biosafety:</b>	1
<b>Culture Medium:</b>	RPMI 1640 + Ham's F12 (1:1) + 2mM Glutamine + 2% Fetal Bovine Serum (FBS).
<b>Subculturing:</b>	Split sub-confluent cultures (70-80%) using 0.25% trypsin or trypsin/EDTA; 5% CO <sub>2</sub> ; 37°C
<b>Subculturing Medium Renewal:</b>	Every 2-6 days
<b>Subculturing Subcultivation Ratio:</b>	1:10 i.e. seeding at 1x10,000 cells/cm <sup>2</sup>
<b>Culture Conditions:</b>	Atmosphere: air, 95%; carbon dioxide (CO <sub>2</sub> ), 5% Temperature: 37°C
<b>Cryopreservation:</b>	95% FBS + 5% DMSO (Dimethyl sulfoxide)

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**SAFETY PRECAUTION:** It is strongly recommended to always wear protective gloves, clothing, and a full-face mask when handling frozen vials. Some vials may leak when submerged in liquid nitrogen, allowing nitrogen to slowly enter the vial. Upon thawing, the conversion of liquid nitrogen back to its gas phase may cause the vial to explode or eject its cap with significant force, creating flying debris.

1. Thaw the vial by gently agitating it in a 37°C water bath. To minimize contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 minutes).
2. Remove the vial from the water bath as soon as its contents are thawed and decontaminate it by dipping in or spraying with 70% ethanol. From this point, all operations must be performed under strict aseptic conditions.
3. For cells sensitive to DMSO, it is recommended to remove the cryoprotective agent immediately. Transfer the vial contents to a centrifuge tube containing 9.0 mL of complete culture medium and centrifuge at approximately 125 × g for 5 to 7 minutes.
4. Discard the supernatant and resuspend the cell pellet in the recommended complete medium (see specific batch information for the appropriate dilution ratio).
5. Incubate the culture under appropriate atmospheric and temperature conditions (see "Culture Conditions" for this cell line).

### Thawing Frozen Cells:

**NOTE:** It is important to avoid excessive alkalinity of the medium during cell recovery. To minimize this risk, it is recommended to place the culture vessel containing the growth medium in the incubator for at least 15 minutes before adding the vial contents. This allows the medium to stabilize at its normal pH (7.0 to 7.6).

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