| | |
|---|---|
| **BCRJ Code:** | 0190 |
| **Cell Line:** | NGM |
| **Species:** | Homo sapiens |
| **Vulgar Name:** | Human |
| **Tissue:** | Skin |
| **Cell Type:** | Melanocyte |
| **Disease:** | Neavo |
| **Growth Properties:** | Adherent |
| **Sex:** | Female |
| **Age/Ethinicity:** | 1 Year / |
| **Derivation:** | This primary human cell was stablished by tripsin digestion of blue neavo skin from 1 year patient undergoing plastic reparative surgery. |
| **DNA Profile:** | Amelogenin: X, X CSF1PO: 12, 11 D13S317: 12, 8 D16S539: 12 D5S818:12, 11 D7S820: 10 THO1: 7 TPOX: 11, 8 vWA: 17, 15 |
| **Biosafety:** | 1 |
| **Addtional Info:** | This cell population apers to be very heterogeneous, containing some keratinocytes also. |
| **Culture Medium:** | 1:1 mixture of Dulbecco's modified Eagle's medium and Ham's F12 medium containing 1.2 g/L sodium bicarbonate, 2.5 mM L-glutamine, and 0.5 mM sodium pyruvate supplemented, 20%(v/v) Fetal Calf Serum, 1,4 uM Hidrocortisone, 1nM Triiodotreonin (T3); 10 ug/mL Insulin, 10 ug/mL Transferrin and 10ng/mL Epidermal Growth Factor. |

| | |
|---|---|
| **Subculturing:** | Remove medium, and rinse with PBS without calcium and magnesium. Remove the solution and add an additional 1 to 2 mL of trypsin-EDTA solution. Allow the flask to sit at room temperature (or at 37°C) until the cells detach. Add fresh culture medium, aspirate and dispense into new culture flasks. NOTE: For more information on enzymatic dissociation and subculturing of cell lines consult Chapter 12 in Culture of Animal Cells, a manual of Basic Technique by R. Ian Freshney, 6th edition, published by Alan R. Liss, N.Y., 2010. |
| **Subculturing Medium Renewal:** | Twice per week |
| **Culture Conditions:** | Atmosphere: air, 95%; carbon dioxide (CO2), 5% Temperature: 37°C |
| **Cryopreservation:** | 95% FBS + 5% DMSO (Dimethyl sulfoxide) |
| **Thawing Frozen Cells:** | 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.<br>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).<br>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.<br>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.<br>4. Discard the supernatant and resuspend the cell pellet in the recommended complete medium (see specific batch information for the appropriate dilution ratio).<br>5. Incubate the culture under appropriate atmospheric and temperature conditions (see "Culture Conditions" for this cell line).<br><br>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). |

| | |
|---|---|
| **Depositors:** | Flávio H. P. Braga; Banco de Células do Rio de Janeiro; Programa Avançado de Biologia Celular Aplicado à Medicina - PABCAM; Hospital Universitário Clementino Fraga Filho - HUCFF; UFRJ |
| **Cellosaurus:** | CVCL_0R16 |