Code:	0190
Cell Line:	NGM
Species:	Homo sapiens **Vulgar Name:** Human
Tissue:	Skin
Disease:	Neavo
Growth Properties:	Adherent
Age Ethinicy:	1YEAR OLD
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
Biosafey:	1
Additional 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, 15 mM HEPES 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. **Medium Renewal:** Twice per week **Subcultivation ratio:**
Culture Conditions:	Atmosphere: air, 95%; carbon dioxide (CO2), 5% Temperature: 37°C
Cryopreservation:	95% FBS + 5% DMSO (Dimethyl sulfoxide)
Thawing Frozen Cells:	SAFETY PRECAUTION: Is highly recommend that protective gloves and clothing always be used and a full face mask always be worn when handling frozen vials. It is important to note that some vials leak when submersed in liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vessel exploding or blowing off its cap with dangerous force creating flying debris. 1. Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the Oring and cap out of the water. Thawing should be rapid (approximately 2 minutes). 2. Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions. 3. For cells that are sensitive to DMSO is recommended that the cryoprotective agent be removed immediately. Transfer the vial contents to a centrifuge tube containing 9.0 mL complete culture medium and spin at approximately 125 x g for 5 to 7 minutes. 4.Discard the supernatant and Resuspend cell pellet with the recommended complete medium (see the specific batch information for the culture recommended dilution ratio). 5. Incubate the culture in a appropriate atmosphere and temperature (see "Culture Conditions" for this cell line). NOTE: It is important to avoid excessive alkalinity of the medium during recovery of the cells. It is suggested that, prior to the addition of the vial contents, the culture vessel containing the growth medium be placed into the incubator for at least 15 minutes to allow the medium to reach 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