Data Sheet

BCRJ Code:	0220
Cell Line:	SF 181
Species:	Homo sapiens
Vulgar Name:	Human
Tissue:	Skin
Cell Type:	Fibroblast
Morphology:	Fibroblast
Disease:	Epidermolysis Bullosa
Growth Properties:	Adherent
Sex:	Male
Age/Ethinicity:	10 Year / Caucasian
Derivation:	Human primary cell line isolated from skin biopsies. Isolated from apatient with epidermolysis bullosa.
DNA Profile:	Amelogenin: X,Y CSF1PO: 10,11 D13S317: 11,12 D16S539: 13 D5S818: 10,13 D7S820: 10,11 THO1: 6,9 TPOX: 11,8 vWA: 16 D2S1338: 19,20 D19S433: 14 FGA: 20,24 D3S1358: 15 D18S51: 13 D8S1179: 13 D21S11: 31,28
Biosafety:	1
Addtional Info:	Formation of blisters and cutaneous lesions upon light mechanical trauma. The byopsia was taken from one of this lesions. Digestive tract also involved, with severe impairement of uper digestive tract function. Patient followed by Dr. Flavio Fraga and Dr. Gisele Pires, Ambulatorio de Imunologia, Hospital Universitario, UFRJ, Rio de Janeiro. Migration assays on collagen gels show a normal phenotype.

Culture Medium:	Dulbecco's Modified Eagle's Medium (DMEM) modified to contain 4 mM L-glutamine, 4500 mg/L glucose and 10% of fetal bovine serum.
Subculturing:	Volumes used in this protocol are for 75 cm2 flask; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes. 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:	2 to 3 times 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: 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:	Marcelo Carvalho, Banco de Celulas do Rio de Janeiro.
Cellosaurus:	CVCL_D682