BCRJ Code:	0368
Cell Line:	A-72
Species:	Canis familiaris
Vulgar Name:	Dog
Tissue:	Unknown
Cell Type:	Fibroblast
Morphology:	Fibroblast
Disease:	Tumor
Growth Properties:	Adherent
Sex:	Female
Age/Ethinicity:	8 Year /
Derivation:	The line was established from a 1 cm diameter tumor taken from the left thigh of a female Golden Retriever.
Virus Succeptility::	Canine adenovirus 2 Canine coronavirus , Canine coronavirus Canine parainfluenza virus Canid herpesvirus 1 Canine distemper virus Canine minute virus
Biosafety:	1
Culture Medium:	Leibovitz's L-15 Medium contains 2 mM L-glutamine and no sodium bicarbonate and fetal bovine serum to a final concentration of 10%. Note: The L-15 medium formulation was devised for use in a free gas exchange with atmospheric air. A CO2 and air mixture is detrimental to cells when using this medium for cultivation.

Subculturing:	Remove medium, rinse with fresh 0.25% trypsin, 0.53 mM EDTA solution, remove trypsin and let the culture sit at 37°C for 10 to 15 minutes. Add fresh medium, aspirate and dispense into new 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
Subculturing Subcultivation Ratio:	1:2 to 1:4
Culture Conditions:	Atmosphere: air, 100% 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).

References:	Binn LN, et al. Establishment of a canine cell line: derivation, characterization, and viral spectrum. Am. J. Vet. Res. 41: 855-860, 1980. PubMed: 6254399 The line was established from a 1 cm diameter tumor taken from the left thigh of a female Golden Retriever. Standard Quantitative Disk Carrier Test Method for Determining the Bactericidal, Virucidal, Fungicidal, Mycobactericidal and Sporicidal Activities of Liquid Chemical Germicides. West Conshohocken, PA:ASTM International;ASTM Standard Test Method E 2197-02.
Depositors:	Ouro Fino Saúde Animal LTDA
ATCC:	CRL-1542