Designation: **B-LCL-HROC43**

Synonym: Bc HROC43

CLS order number: Cryovial: 302067
gDNA: 302067GD5, 5 µg
Snap-frozen cell pellet: 302067CP

Origin and General Characteristics		
Depositor:	Michael Linnebacher	
Organism:	Homo sapiens (human)	
Ethnicity:	Caucasian	
Age:	72 years old	
Gender:	Male	
Tissue:	Peripheral blood from a patient suffering from CRC	
Morphology:	Round cells in cluster	
Cell type:	B lymphoblastoid, immortalized using EBV	
Growth Properties:	Suspension	
Description:	This is one cell line of a series of tumor cell lines which have been established by PD Dr. Michael Linnebacher and coworkers from the peripheral blood of patients suffering from CRC since 2006.	
References:	--	
Culture Conditions and Handling		
Culture Medium:	RPMI 1640 supplemented with L-glutamine and 10% fetal bovine serum.	
Subculturing:	Take an aliquot of the cell suspension to determine the cell concentration, then carefully resuspend the cells and dispense into new flasks which contain fresh medium.	
Split Ratio:	Inoculate the fresh medium with 5x105 cells/ml	
Fluid Renewal:	1 to 2 times weekly	
Doubling time:	n.d.	
Freeze Medium:	CM-ACF (CLS order number 800650, 50ml), serum free, animal-component free	
Freezing recovery:	Fast	
Sterility:	Mycoplasma specific PCR: negative. Mycoplasma specific PlasmoTest: negative. Bacteria, fungi: negative.	
Biosafety Level:	2 According to the German Law for the Protection against Infections (Infektionsschutzgesetz IfSG), cell lines immortalized with EBV fall under Risk group L2, and can only be distributed to customers holding a valid permit of the respective authority (IfSG §44 and §45)	
Safety precautions:	If the cryovial is planned to be stored in liquid nitrogen and to be thawed in the future, special safety precautions should be followed: Protective gloves and clothing should be used and a facemask or safety goggles must be worn when transferring frozen samples into or removing from the liquid nitrogen tank. The removal of a cryovial from liquid nitrogen may result in the explosion of the frozen vial creating flying fragments. Caputo, J.L. Biosafety procedures in cell culture. J. Tissue Cult. Methods 11:223-227, 1988. ATCC Quality Control Methods for Cell Lines, 2nd edition, 1992.	
Special Features of the Cell Line		
Viruses:	EBV+; free of human pathogenic viruses SV40, JC/BK, HBV, HCV, HIV.	
Immunophenotype:	$CD19^+$	
DNA Profile (STR):	Amelogenin: X (Patient male, Y lost) CSF1PO: 11 D13S317: 8,12	D7S820: 13 THO1: 10 TPOX: 11

	D16S539: 9,13 D5S818: 11,12	vWA: 15,17 D21S11: 31.2
	Deletions in the Y-chromosome sometimes cause a misidentification of the biological sample as female. However the cell line is still called authenticated.	
HLA-typing:	Class Ia HLA-A: A*01:01:01,*02:01:01 HLA-B: B*15:01:01,*37:01:01 HLA-C: C*03:03:01,*06:02:01 Class Ib E*01:01:01	Class II DR: DRB1*10:01:01,*12:01:01 DQ: DQA1*01:05:01,*05:05:01 DQ: DQB1*03:01:01,*05:01:01 DP: DPB1*01:01:01,*05:01:01
Related Cell Lines:	**HROC43:** CLS catalog-no **300823**, cryovial; **330823** proliferating culture.	

Certificate of Analysis:	The Certificate of Analysis for each batch can be requested by e-mail at service@clsgmbh.de.

Recommendations for handling of adherent cell cultures following delivery	
Cryopreserved cells	The cells come deep-frozen shipped on dry ice. Please make sure that the vial is still frozen. If immediate culturing is not intended, the cryovial(s) must be stored below -150°C after arrival. If immediate culturing is intended, please follow these instructions: Quickly thaw by rapid agitation in a 37°C water bath within 40-60 seconds. The water bath should have clean water containing an antimicrobial agent. As soon as the sample has thawed, remove the cryovial from the water bath. Note: A small ice clump should still remain and the vial should still be cold. From now on, all operations should be carried out under aseptic conditions. Transfer the cryovial to a sterile flow cabinet and wipe with 70% alcohol. Carefully open the vial and transfer the cell suspension into a 15 ml centrifuge tube containing 8 ml of culture medium (room temperature). Resuspend the cells carefully. Centrifuge at 300xg for 3 min and discard the supernatant. The centrifugation step may be omitted, but in this case the remains of the freeze medium have to be removed 24 hours later. Resuspend the cells carefully in 10ml fresh cell culture medium and transfer them into two T25 cell culture flasks. All further steps are described in the Subculture section.
Proliferating Cultures	Not available as vital cell culture.

Warranty:	CLS warrants for a high cell viability and culture performance only if the product(s) is (are) stored and cultured according to the information described above. Using cell culture media and supplements other than the ones recommended in this product information may result in satisfactory proliferation and viabilities. CLS, however, does not warrant for cell recovery, proliferation and function if differing formulations are employed.
Disclaimer:	The customer shall not be entitled to employ this product for purposes other than research. Commercial utilization shall not be permitted; in particular, the cell line, its components or materials made therefrom shall not be sold or transferred to any third party. In addition, the term ‘Commercial use’ shall mean any activity by a party for consideration and may include, but is not limited to, use of the product or its components in manufacturing, for providing services, e.g. fee for service testing, in quality control or assurance processes within the manufacturing of products for sale, for therapeutic, diagnostic or prophylactic purposes, or for resale. This product from CLS has been manufactured under license for third parties. The customer shall take the submitted licensing terms and conditions of third parties into consideration and may only make use of such within the scope of the rights granted therein.