Designation: **NFS-60**

CLS order number: Cryovial: 400301
Vital: 440301

Origin and General Characteristics	
Organism:	Mus musculus (mouse)
Tissue:	Blood
Morphology:	Lymphoblast
Cell type:	Leukemia, myeloid
Growth Properties:	Suspension
Description:	A murine myeloblastic cell line established from leukemic cells obtained after infection of (NFS X DBA/2) F1 adult mice with Cas Br-M murine leukemia virus. NFS-60 cells are dependent on IL3 for growth and maintenance of viability *in vitro*. These cells are used to assay murine and human G-CSF. This bipotential murine hematopoietic cell line is responsive to IL-3, GM-CSF, G-CSF, and erythropoietin.
References:	Weinstein Y et al. Truncation of the c-myb gene by a retroviral integration in an interleukin 3-dependent myeloid leukemia cell line. Proc Natl Acad Sci USA 83: 5010-4, 1986.
Culture Conditions and Handling	
Culture Medium:	RPMI 1640 medium supplemented with L-glutamine, 1 mM Na-pyruvate, 10% fetal bovine serum and 33 IU/ml mIL-3. As source of cytokines, CLS-conditioned medium supplement (KMG-2, CLS order number 810210, 25ml, 810250, 50ml), 10-20% in regular culture medium may be used as an alternative. The Ready-to-use medium, MG-71, incl. KMG-2, is available by the CLS order number 820701.
Subculturing:	Subculture by transferring an appropriate amount of the cell suspension into new cell culture flasks already containing fresh cell culture media.
Seeding density:	Start cultures at 5 x 10^4 viable cells/ml.
Freeze Medium:	CM-1 (CLS order number: 800125, 25ml, 800150, 50ml)
Sterility:	Fluorescence (DAPI) test: negative; Mycoplasma specific PCR: negative; Bacteria specific PCR: negative
Biosafety Level:	1
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	
Species identification:	Mouse origin was verified by the PCR technique using the Mouse cox I and Mouse J01420 primer.
Viruses:	SMRV: Negative, as confirmed by Real-Time PCR

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

Recommendations for handling of cells growing in suspension following delivery	
Cryopreserved cells	If immediate culturing is not intended, the cryovial(s) must be stored below -150°C or at least at -80°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	This particular cell line will **not** be delivered as **vital culture**, as they stop proliferating immediately once the concentration of the cytokines drop.

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.