

**ASHCS Cryopreserved Atlantic Salmon Hepatocytes for Suspension Assays
Cell Specification – Certificate of Analysis (CoA)**

Lot ASH200923-2 Pool Batch Release: November 27, 2020

Species: Atlantic salmon (<i>Salmo salar</i>) Supplier: Fish farm Danish Salmon, Hirtshals, DK Age: approx. 1 year		Number and gender of animals: 3, female sexually immature All animals were kept under controlled environmental conditions at "Aquaristikshop" in Schwerin.					
Animal characteristics:							
Donor	1	2	3				
Fish weight [g]	308	213	243				
Liver weight [g]	6.8	5.1	6.9				
Gonad weight [g]	0.45	0.45	0.46				
GSI (gonad weight/fish weight)	0.15	0.21	0.19				
GSI = Gonadosomatic index							
Cryopreservation: Date: Sep 23, 2020 Amount per vial: 10 x 10 ⁶ cells		Thawing: n=1 Post-thaw viability: 96.2 % Post-thaw yield per vial: 3.1 x 10 ⁶ cells Recovery: 31 %					
Viability test on orbital shaker (Eppendorf Thermomixer C, 1000 rpm at 14 °C with 0.5 x 10 ⁶ cells in 0.5 ml L-15 medium with 5 % FCS):							
Time [h]	0	1	2	3	4	5	24
Viability [%]	96.2	98.7	94.9	94.5	98.8	96.1	94.0
Determination of CYP activities in suspension (Eppendorf Thermomixer C, 1000 rpm at 14 °C with 0.5 x 10 ⁶ cells in 0.5 ml L-15 medium with 5 % FCS):							
Assay	Enzyme activities (pmol/min*mg protein) mean ± SD						
Phenacetin-O-deethylase	3.1 ± 0.5						
Bupropion-hydroxylase	8.0 ± 1.4						
Midazolam 1'-hydroxylase	17.2 ± 3.7						

Animal husbandry conditions:

Water temperature (°C)	13.5 ± 0.3
pH	8.2 ± 0.1
NH ₄ (mg/l)	1.9 ± 1.2
NO ₂ (mg/l)	0.5 ± 0.4
NO ₃ (mg/l)	1.5 ± 0.6
Carbonate hardness [°KH]	18.8 ± 0.5
Salinity (‰)	32.3 ± 1.2
Conductivity [µS/cm]	49,500 ± 1,731

Note: For thawing of fish (Atlantic salmon) hepatocytes please follow the respective conditions in our manual "Thawing and Culturing of Cryopreserved Primary Hepatocytes in 2D and Suspension".

Store at -150 °C or in the vapour phase of LN₂.

This product should be considered as potential biohazard. Only intended for *in vitro* use.

Issued by: M. Thiede

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