Instrument-Based Hematopoietic Cell Assays from Preferred Cell Systems™

Characteristics	HALO®	HemoGLO™	HemoFLUOR™	MultiCellGro™	HemoLIGHT™
Uses	For all applications where a standardized and validated assay is needed. Basic and stem cell research. Veterinary research. Cellular therapy. Hematotoxicity screening and testing.	For all hematopoietic viability and cell proliferation applications. Basic and stem cell research; Veterinary research; Cellular therapy	For all hematopoietic viability and cell proliferation applications. Basic and stem cell research; Veterinary research; Cellular therapy	For primitive lympho-hematopoietic stem cell applications. Stem cell research; Veterinary stem cell research; Cellular therapy	For all hematopoietic viability and cell proliferation applications. Basic and stem cell research; Veterinary research; Cellular therapy;
Assay availability:	1-plate kit (HALO®-Tox HT only available with 2- and 4-plates)	1-plate kit	1-plate kit	1-plate kit	1-plate kit
Number of samples:	Depends on number of replicates required(1)	Depends on number of replicates required(1)	Depends on number of replicates required(1)	Depends on number of replicates required(1)	Depends on number of replicates required(1)
Type of assay (2):	Suspension Expansion Culture	Suspension Expansion Culture	Suspension Expansion Culture	Suspension Expansion Culture	Suspension Expansion Culture
Assay format:	96-well plate. (384-well plate option for STEMpredict™ and HALO®-Tox HT upon request3)	96-well plate.	96-well plate.	96-well plate.	96-well plate.
Throughput:	High throughput	High throughput	High-throughput capability	High throughput capability	High throughput capability

Preferred Cell Systems™

Cell functionality detected:	Viability/Cell proliferation/ Cytotoxicity/Cell number	Viability/Cell proliferation/ Cytotoxicity/Cell number	Viability/Cell proliferation/ Cytotoxicity/Cell number	Viability/Cell proliferation/ Cytotoxicity/Cell number	Viability/Cell proliferation/ Cytotoxicity/Cell number
Readout:	Luminescence plate reader[4]	Luminescence plate reader[4]	Fluorescence plate reader[4]	Fluorescence plate reader[4]	Absorbance plate reader[4]
Biochemical principle:	Intracellular ATP, luciferin/luciferase	Intracellular ATP, luciferin/luciferase	Resazurin to resorufin (fluorescent)	Resazurin to resorufin (fluorescent)	Tetrazolium WST-1 reduction to yellow formazan
Wavelength (filters)	None. Glow luminescence	None. Glow luminescence	Excitation: 560nm. Emission: 590nm	Excitation: 560nm. Emission: 590nm	Absorbance at 440nm
Time to develop reaction:	10 minutes	10 minutes	1-4 hours	1-4 hours	1-4 hours
Readout endpoint:	Relative luminescence units (RLU) converted to ATP concentration (μM).	Relative luminescence units (RLU) with optional[5] conversion to ATP concentration (μM).	Relative fluorescence units (RFU)	Relative fluorescence units (RFU)	Relative absorbance units
Subjectivity:	Non-subjective	Non-subjective	Non-subjective	Non-subjective	Non-subjective
Cell types detected:	All proliferating stem and progenitor cells	All proliferating stem and progenitor cells	All proliferating stem and progenitor cells	Primitive lympho-hematopoietic stem cells	All proliferating stem and progenitor cells
Species:	Human; Primate; Horse; Pig; Sheep; Dog; Rat; Mouse	Human; Primate; Horse; Pig; Sheep; Dog; Rat; Mouse	Human; Primate; Horse; Pig; Sheep; Dog; Rat; Mouse	Human; Mouse	Human; Primate; Horse; Pig; Sheep; Dog; Rat; Mouse
Alternative to MethoCult™ equivalent assays:	No	Yes	Yes	No	Yes

Total culture volume/sample Required[6]:	Depends on number of replicates required	Depends on number of replicates required	Depends on number of replicates required	Depends on number of replicates required	Depends on number of replicates required
Number of replicates/ culture:	As many as statistically necessary	As many as statistically necessary	As many as statistically necessary	As many as statistically necessary	As many as statistically necessary
Volume/Culture:	0.1mL (25µL for 384-well plates)	0.1mL (25µL for 384-well plates)	0.1mL (25µL for 384- well plates)	0.1mL (25µL for 384- well plates)	0.1mL (25µL for 384- well plates)
Cell culture time[7]:	4-7 days	4-7 days	4-7 days	4-7 days	4-7 days
Instrument calibration:	Yes[8]	Optional[8]	No	No	No
Standardized assay:	Yes[9]	Optional[9]	No	No	No
Proficiency testing incorporated in assay:	Yes[10]	Optional[10]	No	No	No
Measurement Assurance:	Yes[11]	Optional[11]	No	No	No
Validation[4]:	Yes[12]	Optional[12]	No	No	No

(1) The number of samples tested depends on the number of replicates/sample performed. For example, if 4 replicates/sample are performed, then 24 samples can be analyzed on a single 96-well plate. For 6 replicates, 16 samples. For 8 replicates, 12 replicates. These samples, however, do not have to performed on the same day.
(2) Suspension Expansion Culture™ (SEC™) Technology takes the place of methylcellulose, allowing easier and faster dispensing.
(3) Some HALO® assays, including STEMpredict™ and all HALO®-Tox HT assays for hematotoxicity testing are available with 384-well plates upon request.
(4) Multimode plate readers are also av available that have the capability of luminescence, fluorescence and absorbance.
(5) HemoGLO™ does not include the reagents to calibrate and standardize the assay. However, these reagents are available separately for this assay.
(6) The total culture volume will depend on the number of replicates required. For example, if 6 replicates are required, a total volume of 0.8mL should be prepared.

(7) Culture time depends on the state of fractionation or purification of the source cells, cell population tested and species. In some cases it also depends on the assay itself. For example, STEMpredict™ is a 3-day assay.
(8) Instrument calibration is performed using the ATP controls included with the assay kit or obtained separately for HemoGLO™.
(9) Standardization implies performing a standard curve. ATP standards are included in all HALO® assay kits and can be obtained separately for HemoGLO™..
(10) The inclusion of standards and controls allows the results to be compared to measurement assurance parameters (provided) to ensure the assay is working correctly. This allows proficiency testing to be determined directly prior to measuring samples.
(11) = Measurement assurance parameters are included in all HALO® assays and when standards and controls are purchased separately for HemoGLO™.
(12) A validated assay allows the user to trust the results obtained. This can only be achieved if standards and controls are included. HALO® and HemoGLO™ have been validated. The validation parameters are included for all HALO® assays and included when the optional standardization kit is purchased for HemoGLO™. Standards and controls allow for in-house validation.