|  |  |
| --- | --- |
| Test Performed By: Chunhua Li | Date: 1/22/2025 |

**Sample List**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| # | **DNA #** | **Accession #** | **Sample type** | **DNA Conc. (ng/µl )** | **A260/280** | **QC Result** |
| 1 | NTC |  |  |  |  | Pass / Fail |
| 2 | C00026B | Tru-Q 1 (5% Tier) | DNA | 5.0 |  | Pass / Fail |
| 3 | 25BH-015MP0030 | CMOL DNA 305\_D-1 | Bone Marrow | 63.8 | 1.9 | Pass / Fail |
| 4 | 25BH-015MP0025 | CMOL DNA 305\_D-3 | Bone Marrow | 166.7 | 1.9 | Pass / Fail |
| 5 | 25BH-016MP0005 | CMOL DNA 305\_D-4 | Bone Marrow | 113.2 | 1.9 | Pass / Fail |
| 6 | 25BH-015MP0031 | CMOL DNA 305\_D-5 | Bone Marrow | 23.0 | 2.0 | Pass / Fail |
| 7 | 25BH-016MP0003 | CMOL DNA 305\_D-6 | Bone Marrow | 209.3 | 1.9 | Pass / Fail |
| 8 | 25BH-016MP0015 | CMOL DNA 305\_D-7 | Blood | 100.7 | 1.9 | Pass / Fail |
| 9 | 25BH-016MP0014 | CMOL DNA 305\_D-8 | Blood | 37.7 | 1.9 | Pass / Fail |
| 10 | 25BH-016MP0016 | CMOL DNA 305\_D-9 | Bone Marrow | 32.9 | 2.0 | Pass / Fail |
| 11 | 25BH-016MP0022 | CMOL DNA 305\_E-1 | Bone Marrow | 61.8 | 1.9 | Pass / Fail |
| 12 | 25BH-016MP0018 | CMOL DNA 305\_E-2 | Bone Marrow | 4.2 | 1.7 | Pass / Fail |
| 13 | 25BH-016MP0036 | CMOL DNA 305\_F-2 | Blood | 28.6 | 1.9 | Pass / Fail |
| 14 | 25BH-016MP0029 | CMOL DNA 305\_F-4 | Bone Marrow | 63.1 | 1.9 | Pass / Fail |
| 15 | 25BH-016MP0034 | CMOL DNA 305\_F-5 | Bone Marrow | 5.8 | 1.6 | Pass / Fail |
| 16 | 25BH-016MP0031 | CMOL DNA 305\_F-6 | Blood | 5.8 | 1.8 | Pass / Fail |
| 17 | 25BH-017MP0002 | CMOL DNA 305\_F-7 | Bone Marrow | 25.6 | 1.9 | Pass / Fail |
| 18 | 25BH-017MP0005 | CMOL DNA 305\_F-9 | Bone Marrow | 45.8 | 2.0 | Pass / Fail |

**Notes:**

**Reviewed By:**

**QUALITY CONTROL SUMMARY**

|  |  |
| --- | --- |
| **Quality Control Sample** | **Valid/Invalid (circle one)** |
| Blank wells <0.5 RFU | Valid / Invalid |
| Standard curve R2 >0.90 | Valid / Invalid |

\_\_\_RUN ACCEPTED

\_\_\_RUN REJECTED

## CAUSE of REJECTION:

### IF REJECTED, CORRECTIVE/PREVENTIVE ACTIONS:

#### Corrective/Preventative Action Reviewed and Approved By:

Laboratory Supervisor/Director or Medical Director

**REAGENTS and MATERIALS**

|  |  |  |
| --- | --- | --- |
| **Description** | **Lot No.** | **Expiration Date** |
| Quant-iT 1X dsDNA Assay Kit  (Invitrogen Cat. No. Q33232, stored at +4 °C)   * Working Solution * Standards |  |  |
| 96-well PCR plate, black, G-bottom, chimney well  (Greiner Bio-One, Cat. No. 655096) |  |  |

**EQUIPMENT**

| **Description** | **Serial Number, if necessary** |
| --- | --- |
| Flouroskan, Thermo Scientific | 374-700065 |
| Post-PCR clean hood |  |
| Serological pipettor |  |
| Single-channel microliter pipettor |  |
| Multiple-channel microliter pipettor |  |
| Barrier pipette tips |  |

**Procedure Checklist (check each step as completed)**

**Post-PCR Area Date: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ Temperature of Room: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_**

|  |  |
| --- | --- |
|  | Remove Quant-iT 1X dsDNA HS Assay kit from +4 °C and allow to equilibrate to room temperature and turn on Flouroskan. |
|  | Start SkanIt RE software and open NGS Quant-iT 1X dsDNA HS template.skax file located on the Desktop. |
|  | Save As NGS 25-CM13 Library QC\_\_CL to NGS Quant-iT 1X dsDNA HS Results on Desktop. |
|  | Close NGS Quant-iT 1X dsDNA HS template.skax. Do not alter this file in any way. |
|  | Click Plate Layout, name Plate as NGS 25-CM13 Library QC\_\_CL, then click Unknown. |
|  | Click Replicates and change Column to 2. Change Dilution Factor to 1:0.5. |
|  | Using the pointer, select the first well of each Unknown DNA duplicate according to Table 1. |
|  | Select all replicates of each Unknown sample, right-click to Edit Samples and rename according to Table 1. Verify correct positioning of samples in plate. |
|  | Go to Post-PCR room and prepare black microplate and reservoir. |
|  | Add 10 μL of each Quant-iT 1X dsDNA HS Standard to duplicate wells according to Table 1. |
|  | Add 2 μL of each Unknown DNA sample to duplicate wells according to Table 1. |
|  | Add 11 mL Working Solution to reservoir. |
|  | Load 200 μL of Quant-iT 1X dsDNA HS working solution into each microplate well containing Standard or Unknown sample using a multichannel pipettor. |
|  | Tap microplate gently to settle any liquid remaining on well walls. |
|  | Load uncovered microplate into Flouroskan instrument by clicking red Eject button. |
|  | Click green Start button. |
|  | After run is complete, discard microplate. |
|  | Save run and open Notepad. |
|  | Click Standard Curve 1 and verify R2 is >0.90. |
|  | Click Average, SD, CV% 1. Click List. |
|  | Select all rows using Ctrl-A. Right click to Copy selected. Paste to Notepad. Verify sample order is correct on .txt file. |
|  | Save Notepad as NGS 25-CM13 Library QC\_\_CL Results to NGS Quant-iT 1X dsDNA HS Results on Desktop. Copy file to jump drive. |
|  | Close software and turn off Fluoroskan. |
|  | Open file M44i.0 – STEP 2 Library QC by NGS Quant-iT 1X dsDNA HS Data Analysis and save as NGS 25-CM13 Library QC\_\_CL Data Analysis to run folder. |
|  | Open NGS 25-CM13 Library QC\_\_CL Results.txt file and save copy to run folder. Copy values to cell A1 in Raw Data tab of NGS 25-CM13 Library QC\_\_CL Data Analysis file. |
|  | Type NGS 25-CM13 Library QC\_\_CL in cell D1 and technician initials to cell J1 of Results tab. |
|  | Print Results tab and save as NGS 25-CM13 Library QC\_\_CL Data Analysis file. |
|  | Proceed to Step 3. |

**Table 1: Step 2 Layout**

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 |
| A | **Blank** | **Blank** | NTC | NTC | 25BH-016MP0014 | 25BH-016MP0014 | 25BH-017MP0002 | 25BH-017MP0002 |  |  |  |  |
| B | **Std B** | **Std B** | C00026B | C00026B | 25BH-016MP0016 | 25BH-016MP0016 | 25BH-017MP0005 | 25BH-017MP0005 |  |  |  |  |
| C | **Std C** | **Std C** | 25BH-015MP0030 | 25BH-015MP0030 | 25BH-016MP0022 | 25BH-016MP0022 |  |  |  |  |  |  |
| D | **Std D** | **Std D** | 25BH-015MP0025 | 25BH-015MP0025 | 25BH-016MP0018 | 25BH-016MP0018 |  |  |  |  |  |  |
| E | **Std E** | **Std E** | 25BH-016MP0005 | 25BH-016MP0005 | 25BH-016MP0036 | 25BH-016MP0036 |  |  |  |  |  |  |
| F | **Std F** | **Std F** | 25BH-015MP0031 | 25BH-015MP0031 | 25BH-016MP0029 | 25BH-016MP0029 |  |  |  |  |  |  |
| G | **Std G** | **Std G** | 25BH-016MP0003 | 25BH-016MP0003 | 25BH-016MP0034 | 25BH-016MP0034 |  |  |  |  |  |  |
| H | **Std H** | **Std H** | 25BH-016MP0015 | 25BH-016MP0015 | 25BH-016MP0031 | 25BH-016MP0031 |  |  |  |  |  |  |