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SPARC - Attune NxT Set-up for Milli-metabolic bead assay Acquisition

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ABSTRACT

This protocol is to demonstrate how to run a standard plate-based assay on the flow cytometer to produced appropiate concentations of the various analytes to be measured.

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PROTOCOL CITATION

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KEYWORDS

flow cytometry method, fluorescence detection, bead assay, hormone assay

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OWNERSHIP HISTORY

Dec 19, 2019 J Paul Robinson

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31158

GUIDELINES

This assays is specifally designed for the ATTUNE NxT flow cytometer and the Milli-metabolic bead assays

MATERIALS TEXT

MATERIALS

Attune Performance Tracking Beads **Thermo Scientific Catalog #4449754**

Attune Focusing Fluid 1x10L **Thermo Scientific Catalog #A2490**

Attune Wash Solution **Thermo Scientific Catalog #A24974**

Atttune 1x Shutdown Solution **Thermo Scientific Catalog #A24775**

12x75mm test tube **Sarstedt Catalog #55.476.300**

BEFORE STARTING

Setting up instruments correctly is a critical step in any assay

1 Check the waste and

If the waste is full, disconnect the tank and dispose of the waste down the sink with running water. Add **200 mL** of 5% bleach to the tank and reconnect to the Attune NxT. Fill the Focusing Fluid tank from the focusing fluid cube.

2 Check and fill as needed the shutdown solution reservoir and the wash solution reservoir.

3 Also check the waste and focusing fluid reservoirs at the plate sampler station.

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4 At the workstation, click on instrument > start-up at the top of the screen.

5 When start-up is complete, the indicator lights at the instrument will be green.

6 Run the performance test.

7 When the plate finishes, right click on the experiment.

8 Select export FCS files

9 Copy files onto the appropriate directory

10 Analyze data in Mplex software

11 Once data files are analyzed in MPLEX, follow the MPLEX protocol to send data for depositing to NIH. See this protocol for details:

SPARC - Analysis of multiplexed bead data using MPLEX software
by J Paul Robinson

PREVIEW RUN

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