**Agilent Bioanalyzer: Instructions for Supplied RNA 6000 Nano Assay Kit**

Updated: 05/1/2015

Items needed (\*you provide):

Size Ladder 1.2 uL aliquot in 0.5mL clear tube

Marker aliquot in 0.5 mL clear tube

Filtered 32.5 uL Gel Matrix in 0.5 mL clear tube Dye (light sensitive) aliquot in 200uL yellow tube Control Total RNA (supplied upon request, assures assay accuracy)

Nanochip

Centrifuge

Heat block @ 65°C

RNAse Zap & DEPC Water & 1000 ul pipette and tips

\*Gloves

\*10 ul pipette & tips

\*Samples (n= 12 max/nanochip)

5-500 ng/ul, 0.5 mL tubes with 1.5-2.0 uL of sample in ice

(not dry ice)

\*Timer

If purchasing reagents for this kit, the Core maintains aliquots of ladder, marker, filtered gel matrix and dye.

If you have any problems, questions please find Emily or Oanh (754-9143).

\* Set heat block for 65°C.

\* Switch on Bioanalyser (in back). The ‘2100 expert’ software is kept open as it takes a long time to start. However, if it is close please launch it. Verify communication between instrument and computer. Select assay tab, then electrophoresis, then RNA, then Eukaryote or Prokaryote Total RNA **Nano Series II**.

\* Verify Vortexer is functioning.

\* Thaw ladder, vortex, spin down, place on ice. (After denaturing you may need to briefly spin down to collect all liquid to obtain ~1 ul.)

\* Equilibrate marker to RT.

\* Spin down samples briefly if needed to collect liquid at bottom of tube.

\* Equilibrate filtered gel and dye to RT. Add 0.5 ul dye to 32.5 gel ul of filtered gel matrix, vortex, spin for 10 mins. Protect dye and gel-dye mix from light.

\* Denature samples that you already have aliquots of 1.5-2.0 ul in 0.5 mL tubes and aliquot of ladder at 65-

70°C for 2 mins. Spin down. Place on ice.

\* Decontaminate electrodes following instructions in manual on page 16 and keep cleaning chips until the end for use again.

\* **Follow instruction manual on page 20 for assay.** Important to pipette reagents/samples into bottom of well

to avoid bubbles. Don’t forget to vortex nanochip for 1 min at 2400 rpm.

\* Turn off heat block.

\* Clean electrodes with DEPC-water chip, see P29 of manual.

\* Turn off Bioanlayzer.

\* To save data, go to print and save as pdf file, make sure no spaces are in the file name, use underscore instead between words. All data must be dragged over to server by core personnel, please ask anyone who is not working at their bench.

Costs (for UC custumers): Initial training = $109

Nanochip + reagents, you run = $36

Instrument Use: $20