Berthold Microlumat 96V Luminometer Notes (from Manish)

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--Can request cleaning service yearly.
--No lamps.

General Notes

1. Instead of using large plastic dispensers, can use microtubes for reagents in white holder.
2. Glow – slow kinetic – just pipet sample in directly.
3. Flash – Use injectors (e.g. Dual Luciferase Assay)
4. Machine is operated by software, not front panel. Only need to switch on (at back). Since no light, can keep machine on all the time.
5. Uses standard 96well 300 ul white plates. Top read only, so use opaque bottom plates. Can also use half-area plates (170 ul), half diameter
6. There are no gain adjustments on this machine – this is all done automatically.

WinGlow Software

A. LOAD PROTOCOL

I. Wash
Always wash injector tubes (if using) with H20 or ethanol.

II. Select Protocol:
> Integrate – this is an endpoint assay – integrate photon capture over time
> Kinetic – takes lots of readings over a short time span which you define
> Repeated – a SERIES of parallel kinetic assays – reads one well then next

MEASURE
III. Program Injectors:
1. Each well is divided into 3 pies, which indicates each of the 3 PMQ injectors:
> P – Premeasurement injector
> M – Measurement injector
> Q – To stop reaction (quit)

-----Would use P and M for 2 reagents in Dual Luciferase Assay.
2. Highlight each pie accordingly and select volumes to inject
3. Timing – use auto (no need to touch) – set to shortest measuring time. But one could also delay the start of measuring.

IV Program Temperature
Have control over this. Temp reading measures chamber temp, not plate temperature, so advised to place plate in chamber 5-10 minutes before to heat plate.

Luminometer Instructions (cont'd)

V. Repeated Value
1. This indicates how long you wish to measure for.
2. Enter in total time, and the machine will keep re-reading all wells in this time.
3. Cycle time is the interval time, which can adjust (eg. 27sec to 30sec, etc.)
4. The injector head moves in a roster (zig-zag pattern – horizontally,from left to right, then next row below, from the left
5. After this is set, set program the injectors

VI. Kinetic
1. Can set this to DLR (Dual Luciferase) – check kit
2. Measure first reagent for X time , then second reagent for Y time.

VII. Prime
1. This is to get rid of air in injector tubes.
2. Do 3-4 primes and confirm that can see liquid in (spare) microplate.
3. May already be preset.

VIII. After loaded protocol, Press OK.

IX Press Start – will read

X. Export data to Excel if desired.

XI. Recovering Dead Volume (liquid left in injector tubes)
1. Use new plate and return back to Prime Function
2. remove air tubes from liquid (place in open air), so that priming causes forward flushing into new plate
3. Dead volume likely 300-500 ul (recoverable as suggested above)

XII. Wash
Wash injector tubes again with water and ethanol by using priming function until see H20 or ethanol emerge into plate.