**ICCD Camera Technology with Constant Illumination Source in Multiwavelength Analytical Ultracentrifugation: Electronic Supplementary Information**

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ESI Table 1; Performance specification comparison of an Ocean Optics USB2000+ Spectrometer and Xenon flash lamp based spectrophotometer system for MWL AUC detection versus an Andor iStar ICCD Shamrock Spectrograph and Deuterium-Tungsten lamp based spectrophotometer system\(^1\)\(^-\)\(^4\).

| Specification                     | USB2000 Spectrometer / Xenon Flash Lamp                                                                 | Andor iStar / Deuterium-Tungsten Lamp                                                                 |
|-----------------------------------|----------------------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------|
| **Significant Advantages**        | Small, inexpensive, fits in XL-A vacuum chamber                                                        | ICCD allows for gating and signal amplification (both are required for use of a constant illumination source) |
| **Spectral Profile**              | Large emission spikes in Vis, limiting dynamic range;                                                   | Smoother balanced distribution UV through Vis                                                        |
| **Spectral range**                | Dependent on grating, acquisition settings and desired spectral range                                  | UV/Vis, 190 - 780 nm by software selectable grating. Easily exchangeable grating turret.             |
| UV                                | Limited signal at 230 nm, but possible with flash integration.                                         | A-3R2-GRT-0400-0250 UV grating (190 - 400 nm), 400 l/mm, 250 nm blaze, 1.4 nm resolution          |
| **Vis**                           | Excellent visible response up to 650 nm                                                                | A-3R2-GRT-0400-0550 Vis grating (400 - 790 nm), 400 l/mm, 550 nm blaze, 1.4 nm resolution          |
| **NIR**                           | With Xenon Lamp and XR grating 650 nm, up to 800 nm using high sensitivity mode and neglecting visible signal | Not yet tested with additional NIR gratings. The Gen 2 intensifier is limited to 850 nm            |
| **Broad spectra**                 | Not possible to get UV, Vis, and NIR in same acquisition                                               | Dynamically switchable UV/Vis grating options                                                      |
| **Vac Issues**                    | Spectrometer inside; need fibers optical cables                                                        | Spectrometer outside vacuum chamber; mirror optics                                                   |
| **Repetition rate**               | Limited at high rotor speeds by flash lamp repetition rate                                             | Only dependent on rotor speed                                                                       |
| **Acquisition width**             | \(^+\)2 µs, determined by Xe pulse width                                                                | Variable with ns precision; can match channel width of given rotor speed                             |
| **Lamp power**                    | Very high (illumination optics dependent) over short \(^+\)2 µs duration                              | Much lower than Xe lamp (can compensate by acquisition width adjustment and signal integration)    |
| **Detector**                      | Sony ILX511 2048 pixel linear CCD sensor                                                               | Andor iStar A-DH320T-2SF-04 1024x256 pixel intensified CCD, P46 phosphor & 25 mm 2nd generation photocathode (W) |
| **Pixel Size**                    | 14 x 200 µm                                                                                            | 26 x 26 µm                                                                                         |
| **Pixel integration**             | Not possible (can average after in software)                                                          | Software selectable, can set to match spectral resolution and image height                          |
| **Dark noise**                    | Single dark spectrum) 50 counts RMS                                                                    | Negligible with cooled CCD chip                                                                    |
| **Overall DNR**                   | Wavelength and sample concentration dependent (limited by Xenon emission spikes)                      | *not tested                                                                                       |
| **Specified DNR**                 | \(2 \times 10^4\) (system), 1300:1 (single acquisition)                                               | *not specified by manufacturer                                                                  |
| **Pixel well depth**              | *not specified by manufacturer                                                                      | 500,000 e-                                                                                        |
| **Readout rate**                  | 2.4 MHz                                                                                                | 0.05, 1, 3 or 5 MHz (software selectable)                                                          |
| **Read noise**                    | Single dark spectrum) 50 counts RMS                                                                    | Readout rate dependent; @ 1 MHz; 13 e-                                                               |
| **Gain**                          | No gain adjustment option                                                                             | Software selectable                                                                                |
| **Sensitivity**                   | *not specified by manufacturer                                                                       | 2 to 10 e-/count provided in documentation                                                        |
| **Linearity**                     | 0.1 to 1.2 OD                                                                                         | *not tested                                                                                       |
| **SNR**                           | 250 specified by manufacturer                                                                        | Dependent on acquisition settings                                                                  |
| **Illumination**                  | Requires Xenon Flash Lamp                                                                             | Allows for Deuterium-Tungsten Constant Source                                                        |
ESI Figure 1; Illumination optics designs, modeled in Winlen 3D Basic from Qioptiq. Screen capture of Winlen 3D model for focusing Tungsten lamp into Hamamatsu X2D2 Deuterium ‘see-through’ lamp.

ESI Figure 2; Test setup schematic. The 60 mm focal length lens may be either bi-convex for spot-focused illumination, cylindrical for line-focused illumination, or removed entirely for collimated illumination.
ESI Figure 3; Compact imaging optics are designed based off the concept of the mirror imaging optics published previously, and configured to function within the CFA AUC platform. Off axis parabolic mirrors are selected to accommodate the working distance from the CFA detector platform to the rotor holes. The two identical 152.4 mm focal length off axis mirrors from Thorlabs are arranged with confocal geometry. Additional plane mirrors are included to direct the beam along the necessary trajectory of the spectrometer assembly. A sliding stage was designed to hold the spectrometer system and mount onto the optical table of the CFA. The scanning spectrometer track is designed with high precision Schneeburger (4xRN4-225 Rails and 2xKBN4x31 Roller Cage) linear guides, including hardware to mount a Zaber stepper motor (NM17CD-T4- MC04-HSM8 Motor and X-MCB1-KX13B Controller) for radial positioning. The complete system design is shown in ESI Figure 2, and CAD files are available from the authors upon request. (a) Shows the mirror imaging prototype optics constructed with Linos micro-bench rail system parts, two 152.4 mm off-axis parabolic mirrors, and two plane mirrors from Thorlabs. (b) Shows the design concept for the sliding stage to mount an Andor iStar spectrometer system and mirror imaging optics.

ESI Figure 4; The imaging optics prototype performance is demonstrated by a simple imaging test with a Thorlabs 1951 USAF Resolution Test Target, shown on the left. An ideal Image of 1951 USAF test target from Thorlabs is shown on the right. The image was recorded with a Sony XC-EI30 CCD camera. Determining the resolution from the test target requires identifying the line pairs that may be adequately resolved. In the image, Group 4 - Element 5 is the last resolvable line triplet set. This equates to an optical resolution of 19.7 μm, and fits within the
tolerance of the 25 μm spectrometer slit. It is clear from the image that the resolution is better across the vertical dimension, which corresponds to the dimension parallel with the parent parabola of the mirror, and is the dimension most critical for imaging onto the spectrometer entrance slit.

References

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