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An Always-On Remote Flowcell Monitoring Module

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BACKGROUND

For many researchers, particularly those working on cutting-edge research in flow cytometry, the ability to monitor the flowcell can be crucial to the success of your experiments. Many sample or fluidics problems can be easily remedied if the researcher can simply see what is going on inside their flowcell. However, with most flow cytometry instruments, this is difficult or impossible.

METHODS

We have developed a dedicated, always-on flowcell monitoring solution, dubbed the *Cavour*, to address this need. To reduce costs, the *Cavour* is designed using standard off-the-shelf components. The system uses 3D-printed mounts which can be customized, allowing the device to be adapted to most commercial flow cytometry systems (Figure 1). Based on the specific requirements of flow cytometry users, we have implemented a proprietary microscope design which allows remote monitoring of an instrument without removing the cover.

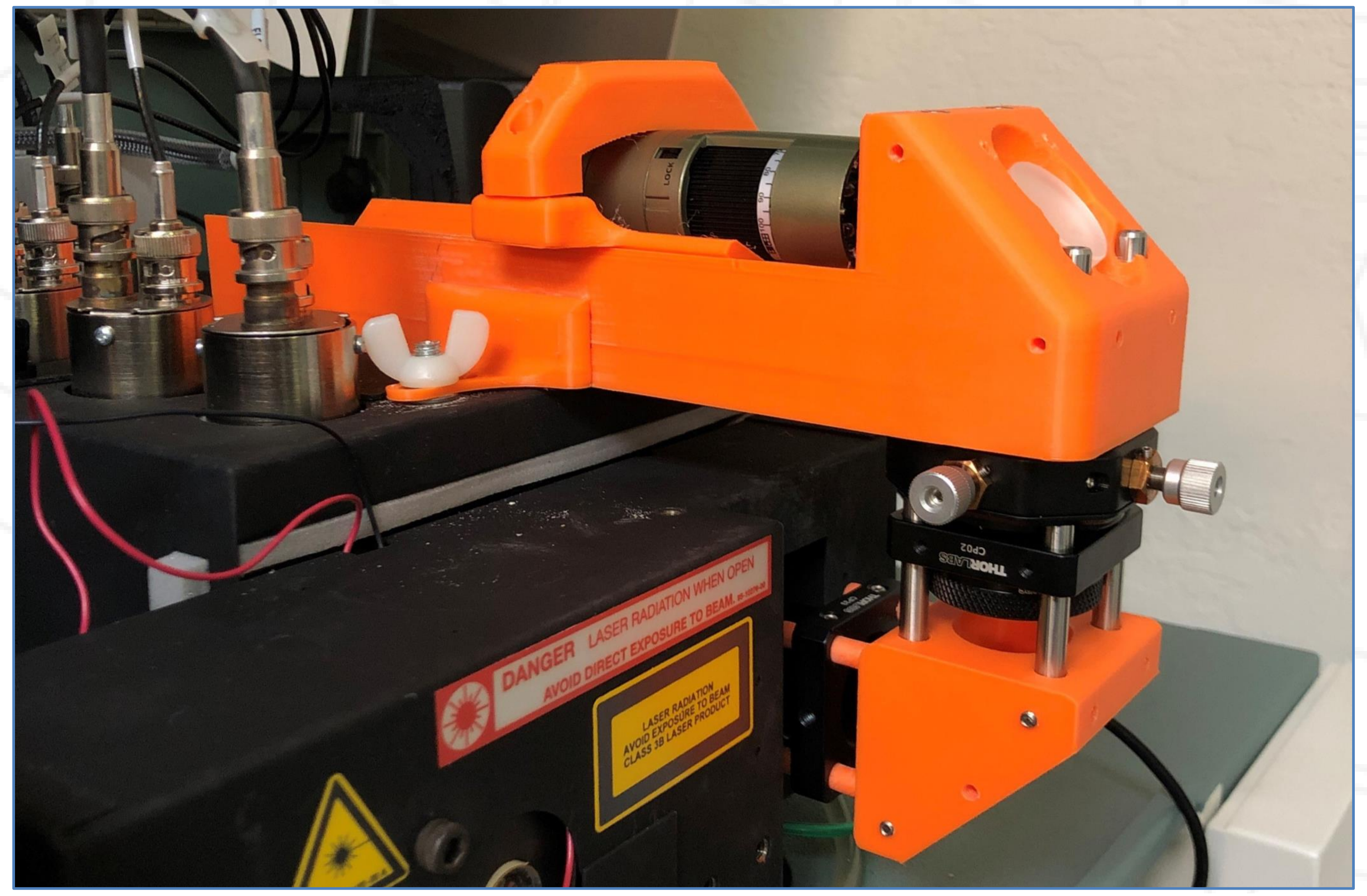

Figure 1. Close-up of the Cavour retrofitted to a flow cytometer using a custom-designed, 3D-printed mount.

Coupling intuitive digital microscope software (DinoCapture) with a tablet provides a continuous live feed of the flowcell and enables always-on monitoring. Our unique optical design provides a large field of view (600 µm x 400 µm) with 50x equivalent magnification, optimized for flowcell viewing. The software allows for image and video capture, and on-screen measurements. The proprietary patent-pending* design allows for adjustment of light collection sensitivity which can be reduced for laser alignment and maximized for imaging and measurements involving microspheres or cell samples.

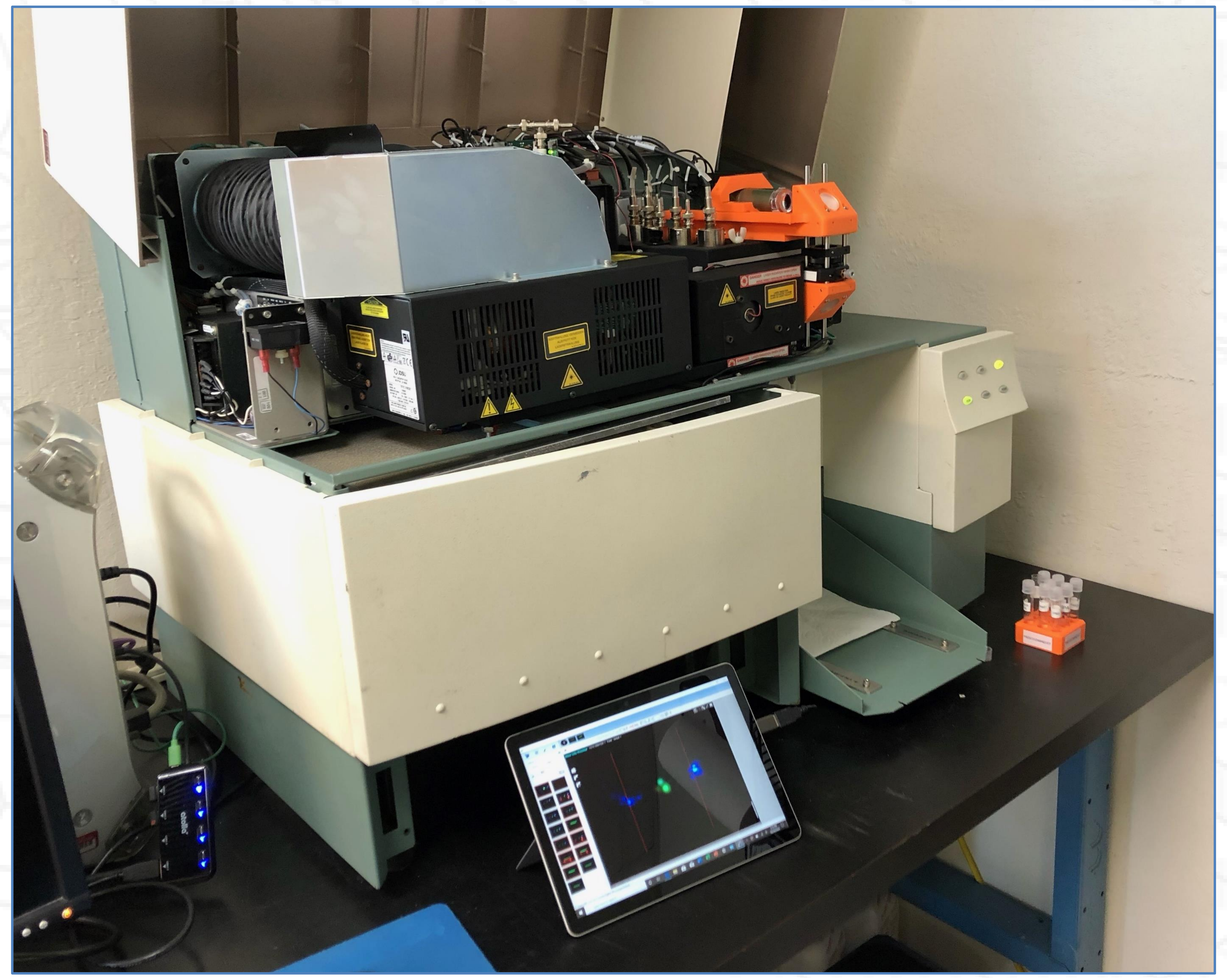

Figure 2. A Cavour retrofitted to a BD FACSCalibur fits completely under the instrument cover. An external tablet can be positioned wherever desired, enabling continuous remote monitoring with the cover on.

We have also incorporated the ability to insert notch filter(s) for laser line(s) in use, improving sensitivity by blocking scattered laser light. These features, based on end-user feedback, are all integrated into the *Cavour*, enabling researchers to perform a variety of troubleshooting activities simply and easily, giving them the ability to address problems on their own without requiring service calls.

RESULTS

The *Cavour* provides many advantages to flow cytometrists. The concept of 3D-printed mounts allows the *Cavour* to be simply integrated into virtually any existing system quickly and at low cost. The *Cavour* is small (10" x 5" x 8") and lightweight (750 g). The *Cavour* allows the user to perform laser alignment and optimization without exposing their eyes to the hazard of looking directly at laser light. Once installed, the *Cavour* allows continuous inspection of the flowcell without ever needing to lift the instrument cover. Figure 2 shows a *Cavour* integrated into a BD FACSCalibur™ instrument, with controls and visualization achieved by an external tablet.

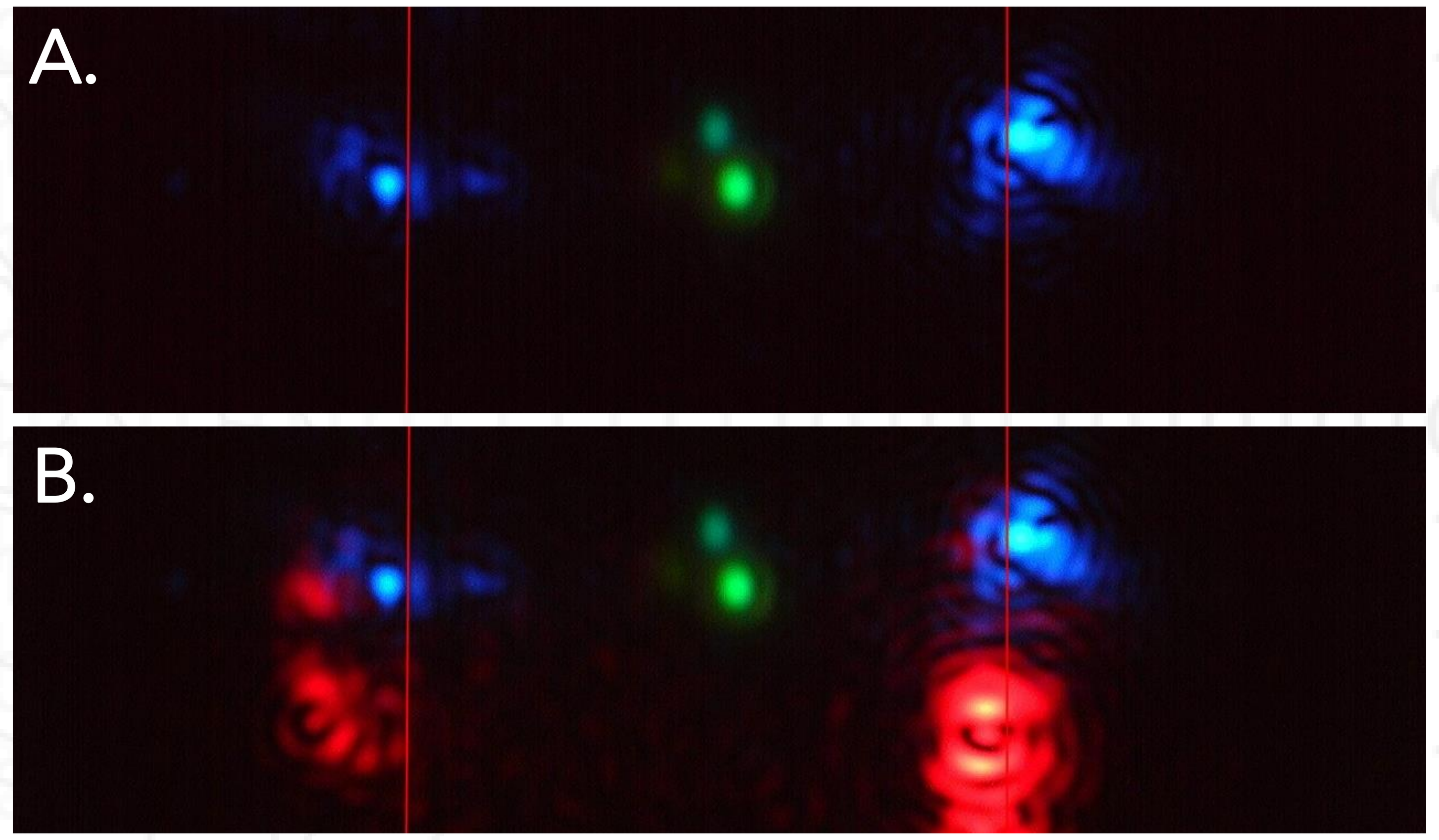

Figure 3. A Cavour image of a properly aligned system with the sample stream (fluorescein) properly confined (green spot) in the flowcell (red lines). Minimal laser scattering by the flowcell walls (A) with 488-nm excitation (blue scattering signal) or (B) with 488- and 635-nm excitation (blue and red scattering signals).

Troubleshooting and optimizing your system's optical alignment, including tasks such as laser realignment (see Figure 3), maximizing signal prior to a key experiment, or identifying deposits in the flowcell that can cause increased background due to light scattering, are all able to be addressed easily, often without having to remove the instrument cover. Researchers are also able to troubleshoot fluidic issues such as bubbles, clogs and debris, and optimize core stream and flow stability. Figure 4 shows diagnosis of low sheath pressure, resulting in a widening of fluorescent signal. On-screen measurements (Figure 5) assist with troubleshooting. The ability to take snapshots and videos allow you to send these diagnostic images to the manufacturer's service technician for more in-depth troubleshooting

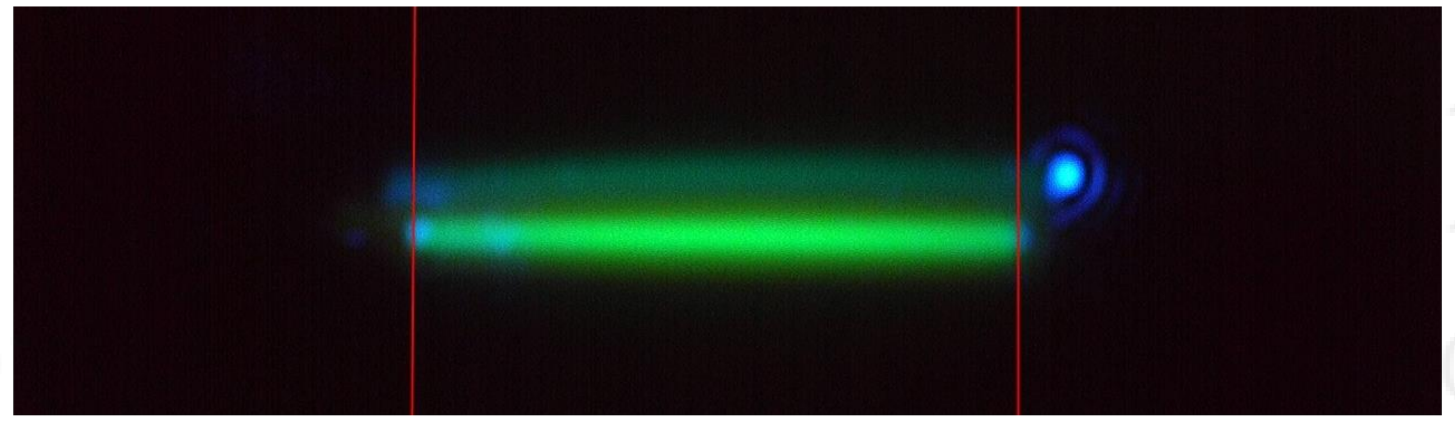

Figure 4. A Cavour is used to diagnose low sheath pressure, with the fluorescein sample (under 488-nm excitation) filling the entire width of the flowcell (red lines).

CONCLUSION

For many flow cytometry researchers, the ability to easily monitor the flowcell and understand what is going on with the core stream is highly desired. The *Cavour* Flowcell Monitoring Module was developed with that in mind. The module is designed to allow it to be adapted to most flow cytometers on the market. Implementing the device should allow researchers to avoid unnecessary service calls and reduce system downtime.

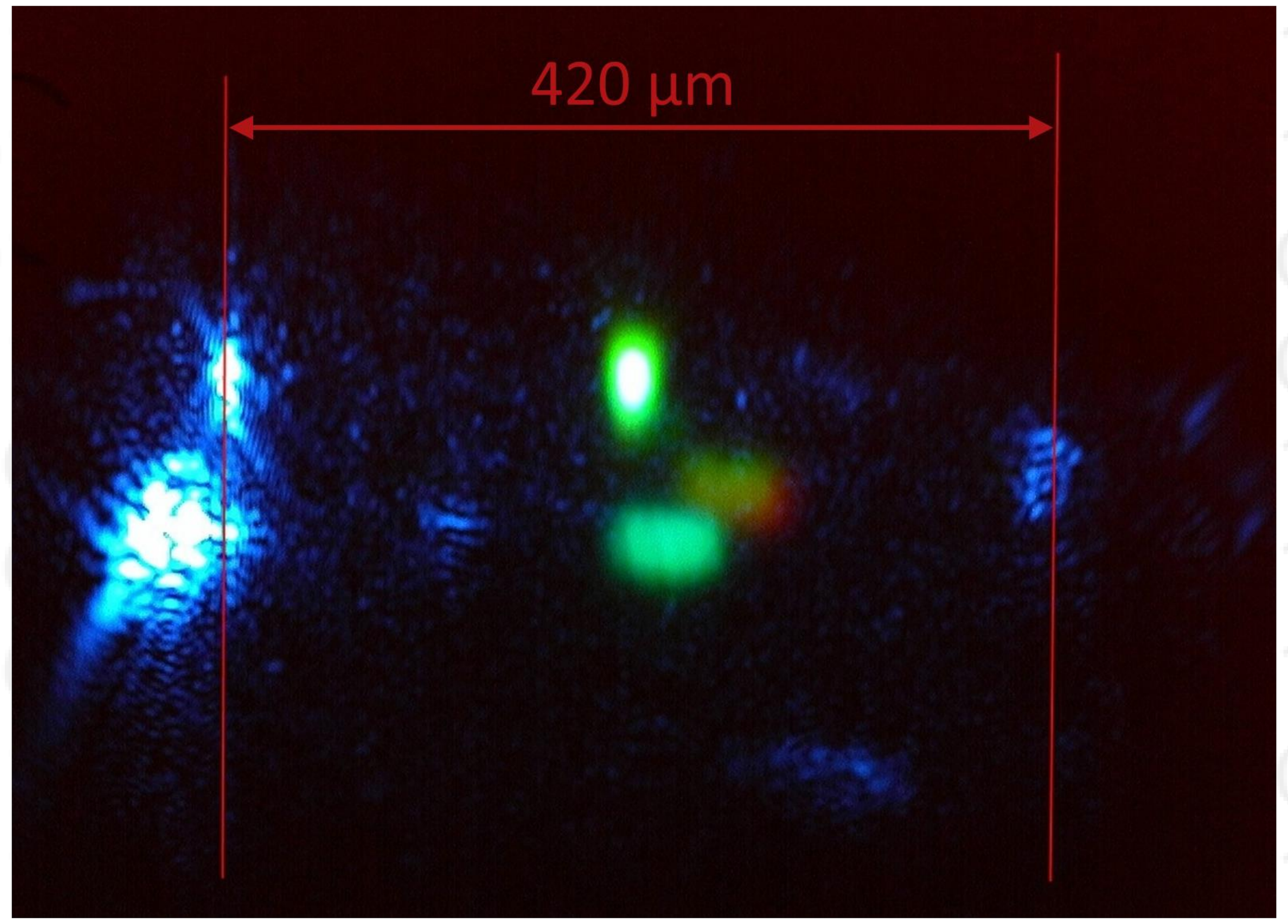

Figure 5. The Cavour software enables on-screen, real-time measurements that simplify troubleshooting.

** The Cavour, or use thereof, may be covered in whole or in part by patents in the U.S. and other jurisdictions. A current list of applicable patents can be found at https://www.kineticriver.com/kinetic-river-corp-patents.*