Automated tools for cell-based screening assays

BY MELISSA FOSHEE AND GREGORY PORTER

As we enter the postgenomics era, it is the information held in the cell that is gaining attention. In order to have a complete picture in drug discovery, it is critical to obtain a perspective that is cell-based and focused on isolated genes or proteins. Information in the cell is what defines the temporal and spatial interactions of the cell's components and, therefore, its normal and abnormal functions. Belief in this concept is growing and has led to research areas that target the whole cell (cellomics) and physiological pathways (physiomics). Research in these new fields has resulted in a significant rise in the use of cell-based assays as screening tools in research and drug discovery.[1]

The scientist's quest for more biologically relevant data from primary screening has produced a dramatic increase in cell-based assays in high-throughput screening (HTS) laboratories. The power of cell-based assays in conjunction with new array and analytical technologies will enable drug discovery processes to be streamlined, reducing the time and cost of moving a lead compound into clinical trial.[2]

Evolution of cell-based assays

Throughout history, biochemical assays have been used with far more frequency than cell-based assays in HTS. These biochemical assays are relatively easy to implement with HTS, but important biological issues are often overlooked when using biochemical assays. These include cellular toxicity and the complex cell biology that surrounds the target compound. Cell-based assays in HTS have helped to fill this void and have enjoyed a rapid rise in popularity.[2] For primary screenings, biochemical assays are still being performed using huge compound libraries. However, cellular assays are being utilized with more frequency for lead selection and lead profiling, with widespread use of HTS technologies for the functional analysis of drug compounds in live cells.[3]

Cell-based screening assays have faced many challenges in the past and were once considered slow, expensive, and lacking reproducibility.[2] Previously, many cell-based assays could not be miniaturized for work in microplates because the readers were not sensitive enough to operate with the low numbers of cells that can be grown in the microplates. In addition, cell culture work is time consuming,and costly; it can occupy more than one-quarter of a laboratory technician's time and requires visits to the laboratory on weekends and holidays.[4]

Automated solutions

The advent of more sensitive detection systems and methods has led to an increase in the development of cell-based assays for screening. This, in turn, has resulted in automated solutions for cell-based assays, helping to reduce the need for human intervention for the repetitive task of cell feeding and also the risk of contamination, which is a common problem with manual cell feeding. Automating the cell feeding process improves speed; reduces cost; and increases reproducibility, ultimately optimizing cell analysis.

Step 1: Automating cell maintenance

Cells need to be fed regularly to survive. This requires that laboratory personnel feed the cells daily, even during inconvenient times. It is a labor-intensive, inconvenient step in the process, yet it is critical when working with cell-based screening assays. However, for the modern laboratory operator, a number of automated solutions are available to simplify cell feeding.

Figure 1 GENios Pro multipurpose reader and dispenser.

The first step in automating cell-based screening is to eliminate the burden of daily cell feeding. The Cell Maintenance System (**Tecan U.S.**, Research Triangle Park, NC) removes the human intervention factor for the monotonous task of cell feeding, reducing the risk of sample contamination. The system comprises the Genesis Freedom workstation (**Tecan**) and its eight tips and robotic manipulator arm (RoMa). The totally robotic automation solution also includes cooled media storage, warmed dispensing stations for up to four different types of media, and a large carousel that contains a humidity-controlled CO_2 incubator. An important feature is the ability of the operator to automate a wide variety of time-consuming tissue culture operations in a completely sterile environment while eliminating routine tissue culture procedures.[4]

A wide variety of additional automation solutions are offered for automated cell-based screening. The Cell Migration Workstation (**Tecan**) utilizes the Genesis Freedom workstation and a multipurpose reader and dispenser such as GENios Pro (**Tecan**) to study the transmigration of cells through a membrane. Many assays can be automated on the Genesis Freedom, including fluorescence, absorbance, luminescence, and fluorescence polarization.

Step 2: Automating assay development

The step following cell maintenance is assay development. The GENios Pro (*Figure 1*) provides a variety of read modes for assay development. This allows scientists to test several detection modes to determine the one that is optimal for their assay. During the development of a screening assay, several types of assays are often tested to determine the one that yields the best sensitivity and uses the least amount of expensive reagent. The ability to miniaturize is vital in scaleup, in which thousands of samples may be run per day. The GENios Pro accommodates low- and high-density plates, and is thus well suited for both development and HTS.

The benefits to using the GENios Pro for assay development are 1) an enhanced optic design that is optimized for cell-based assays, 2) an injector option that pemits one or two injectors for 96-well plates or one for 384-well plates, and 3) dispense volumes ranging from 5 to 350 µL. Injector option features include minimal tubing; very low dead volume, requiring minimal amounts of potentially expensive reagents; a pump-back option that prevents wasting reagents; an injector head that can be removed easily for clean vcumes. In addition, injection parameters can be optimized with the injector head external to the instrument to visualize the force with which the liquid is injecting to determine the optimal speed for injection, which prevents splashing or bubble formation. This allows for optimization without the risk of contaminating the optic head (*Figure 2*). The GENios Pro is compact, with all reagent bottles stored internally, therefore requiring minimal space on or below the deck of the Genesis Freedom workstation. In addition, it can interface with the CONNECT stacker system (**Tecan**), a plug-and-play instrument that offers walkaway batch processing for all microplate formats.

Figure 2 GENios Pro injectors set for external optimization.

Step 3: Automating the cell-based screening process

After assay development, the final step is to automate the cell-based screening process. A wide variety of cell-based screening assays (i.e., calcium flux, cell toxicity, cell proliferation, cell migration, etc.) can be automated using the Genesis Freedom workstation. Cell migration is one example of an assay that uses top and bottom read capabilities, and is found on the GENios, Spectrafluor Plus, Safire, GENios Pro, and Ultra Evolution readers (**Tecan**). The GENios Pro is well suited for calcium flux assays that require injection followed by rapid reading. The readers are appropriate for both assay development and screening assays, leading to a smoother transition from assay development to HTS, since no change in instrumentation is required.

The Genesis Freedom workstation's many configurations allow it to be adapted to virtually any assay with the addition of incubators and shakers. The entire system can be placed within a laminar flow hood for sterility, which is essential to cell-based assays. The workstation can access instruments placed in the cabinet under its worktable, including an automation-friendly centrifuge suitable for working with cell suspensions in microplates. Storage devices and microplates can also be situated beneath the deck. The workstation can be equipped with vacuum devices for work with filter-based assays.

Conclusion

With the increasing need for more biologically relevant data, cell-based assays are a critical tool in drug discovery.[2] As these cell-based screens continue to take precedence over biochemical assays, advances in automation will become essential in the goal of identifying and prioritizing leads for potential drug development candidates. The main advantage of using robots for cell-based screening is that automation removes the human intervention factor, therefore saving time and money and providing improved reproducibility.

References

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Ms. Foshee is an Associate Product Manager, Microplate Systems, and Dr. Porter is a Liquid Handling Product Manager, ***Tecan U.S.****, P.O. Box 13953, Research Triangle Park, NC 27709, U.S.A.; tel.: 800-352-5128; fax: 919-361-5201; e-mail: melissa.foshee@tecan.com.*