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Introduction to Screening Automation

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1. History of Screening Automation

In the late 1970s and early 1980s, the components that have made modern high-throughput screening (HTS) laboratory possible came together. Those were: 1) small scale servo-driven robotic devices; 2) the personal computer; and 3) the microplate. The word "robot" is derived from the Czechoslovakian word Robota, which is translated into English as servant, slave, or laborer. The Robot Institute of America (RIA) defines a robot as a "reprogrammable multifunctional manipulator capable of moving materials, parts, or tools through variable programmed motions for the performance of a variety of tasks" ***(1)***. The technology for servo-controlled robotics was developed in the late 1960s, and employed in the automotive assembly industry. In the 1970s the development of the microprocessor drastically decreased the cost and scale of the control systems for such robotics, making smaller robots feasible. Cartesian robots had three degrees of freedom (XYZ) and were usually mounted in an XYZ frame. Cylindrical robots had four degrees of freedom: rotation at the base and wrist, elevation, and lateral movement, thus defining a workspace similar to a cylinder. Articulating robots added yet another (fifth) degree of freedom, mimicking the human arm with shoulder, elbow, and wrist rotation. The shoulder joint is mounted on a base allowing the entire arm to rotate. The wrist motion has both pitch and roll, allowing complex movements that were desired at the time by the automotive and electronics industry. The Microbot Alpha was an early small articulated robotic arm intended for educational use that sold for about $5000, and was used in 1981 in the first published example of robotic laboratory automation ***(2)***. Zymark Corporation (Hopkinton, MA) created their own cylindrical geometry robotic arm controlled by their proprietary "personal

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computer" and began marketing to the laboratory market in 1982 ***(3)***. Although the first 96-well plastic plate was created in 1952 at the N.I.P.H in Hungary, it wasn't until 1974 that the format was first used for an enzyme-linked immunoserbent assay (ELISA) assay in London and at the Centers for Disease Control (CDC).

1.1. Natural Products Screening

Some of the earliest pharmaceutical screening automation was developed to search natural-product libraries for active compounds. Most, if not all, of this automation was custom-developed in-house and not published due to the competitive nature of the work Thus no common format or technology arose from these efforts. One published case, by scientists at Eli Lilly and Company (Indianapolis, IN) ***(4)***, utilized a PUMA 560 robot to inoculate microbial colonies into vials, leading to a downstream testing of the fermentation extract for antibiotic activity.

1.2. Early Microplate Automation

The first published examples of robotic microplate automation were presented at the Fourth International Symposium on Laboratory Robotics in 1986 ***(5–8)***. All were examples of ELISA automation, and while not focused toward compound screening, contained the roots of today's HTS. Using versions of Zymark's early microplate management system, these were examples of how robotic technology was initially used in the laboratory as a "one-armed chemist" ***(9)*** to anthropomorphically mimic human tasks. Through the use of interchangeable "hands," the robot arm was directly involved in many laboratory unit operations (LUO's) ***(1)*** such as single and multi-channel pipetting, plate washing, reagent dispensing, and pipet tip attachment/detachment. This approach limited throughput to several tens of plates/d. Reliability for unattended operations was moderate due to the many nonideal operations thrust upon the robotic arm.

1.3. Advent of Microplate-Focused Workstations

It was immediately obvious that the core of microplate automation lay in liquid handling, and equally obvious that articulated robotics arms were not the cost-efficient way to automate this process. Several companies created specialized cartesian-geometry liquid-handling robots. Beckman Instruments (Fullerton, CA) developed the most microplate focused device in the Biomek™ 1000, which offered interchangeable single and multichannel pipetting tools and an integral spectrophotometer. Other companies, such as Hamilton Co. (Reno, NV) and Tecan Inc. (Research Triangle Park, NC) developed more general-purpose devices which initially featured single-channel, fixed-tip liquid

handling. Compared to attaching liquid-handling "hands" to articulated robots, these dedicated cartesian geometry systems offered much faster and higher-quality liquid transfer, but a more limited set of LUOs. Thus "workstations" were differentiated as specialized robotic devices focused on a limited set of LUOs vs general purpose robots capable of executing many different LUOs and transporting more than liquids.

The workstation technology immediately found acceptance in early molecular biology/genomics efforts, due to the low-throughput, liquid-handling intensive nature of the work ***(10)***.

1.4. Evolution of the Transport/Workstation Integrated System

In an effort to make integrated, multiple-LUO, general-purpose robotic systems more reliable and capable of higher throughput, system designers began off-loading sample manipulation tasks from the robot arm to more and more specialized workstations. An early example similar in concept to current HTS automation was developed by Beckman Instruments combining their Biomek™ 1000 liquid-handling robot with a modified Zymate arm, renamed the Biomek SideLoader™. This system, described by McRorie for screening large numbers of molecules using a receptor-binding assay ***(11)***, used the robotic arm only for the transport of microplates and pipet tips to and from the Biomek liquid-handler. Later, plate reader and incubator modules were added. This approach represents the realization that general-purpose robotic arms are best at transporting samples, not performing manipulative LUOs. Automated transport of microplates to/from specialized workstations was key in enabling the assay throughput and reliable unattended operation necessary for the evolution of HTS, and remains the basic model for fully integrated assay systems today with variations in the actual transport device. A side effect of parsing out robotic tasks to workstations was a marked increase in the cost of automated systems, reported to be approx $60,000 for an ELISA system in 1986 ***(7)*** and that today can range from $200,000 to well over $1,000,000.

2. Screening Automation Today

The explosive growth of HTS has led to a great abundance of automation technology, ranging from simple, small, and affordable liquid-handling workstations to very large factory-style integrated systems, with a continuum of options in between. Below is an attempt to classify the current types of offerings and their functions.

2.1. Simple Workstations

This category includes small liquid handling robots with fixed single- and multichannel pipetting, priced under $50,000 from several manufacturers

(**Appendix 1**). These are directed toward non-HTS operations such as assay development, and are meant to be "personal" workstations with simple software and small footprints. Also included in this category are other single-task devices such as plate washers and reagent dispensers.

2.2. Standard Workstations

Priced approx from $50,000–150,000, these liquid-handling workstations compare to simple workstations by: 1) larger size, still designed to fit on a laboratory bench (up to 1 meter in length, 30" depth); 2) larger work surface; 3) more feature-rich GUI-style software; 4) variable span and individually addressable multichannel pipetting; 5) washable or disposable pipetting tips; and 6) liquid-level sensing. Workstations in this category may have two rather than one robotic pipetting arm. Some include basic ability to move plates about the worksurface, to/from storage devices such as carousels or stackers as well as remove and replace plate lids. Vendors include Tecan, Beckman, Hamilton, and Packard. Also included in this category are 96- and 384-channel liquid handling workstations, which may have most of the features described earlier with the exception of variable-span and individually addressable pipet tips.

2.3. Extended Workstations

These devices are differentiated from standard workstations by: 1) larger size, as long as 2 meters, but still of a depth to rest on a 30" laboratory bench; 2) full ability to move plates to/from devices such as plate readers, washers, hotels, carousels, incubators, and stackers that are built in or attached to the workstation; 3) dual-pipetting arms, one of which may be configured for 96-channel pipetting; 4) software capable of optimally and automatically scheduling the complex interactions of the workstation's feature set; 5) price: from $150,000 to $300,000. These workstations differ from "Integrated Systems" described in **Subheading 2.4.** in that they are limited in their reconfigurability outside of the initially purchased feature set.

2.4. Integrated Systems: Multitasking Robotic Transport

These systems are descendents of the early robotic ELISA approaches, now with the robotic arm acting only to transport microplates and consumables to/from workstations, and are capable of processing tens of thousands of assays per day. These systems are "multitasking" in that one microplate may visit a given workstation multiple times in the course of an assay. This minimizes the number of workstations needed, requires scheduling software to optimize and control the interleaving of processes, and usually causes the robotic transport to be rate-limiting. A great number of liquid-handling workstations, readers, washers, and other devices are today available for integration into these systems,

with the limitation primarily the variety of devices a system integrator is able to technically support. As the size and number of workstations in an integrated system has grown to generate increased throughput, robotic arms have been mounted on rails to extend their work envelope. Integrated systems may be as large as 3 meters in length and 2 meters in depth operating on both sides of a rail. Systems of this size require significant open space within a laboratory and usually are built on a specialized, stable table structure. These systems are generally very reconfigurable (with appropriate expertise) and adaptable to new devices due to the flexibility of the robotic arm transport in addressing different device designs. The cost of such systems range from $250,000 upwards.

2.5. Integrated Systems: Linear Transport

An alternative to multitasking integrated system transport is a "linear" transport system. In the full implementation of this model, plates only pass through a given workstation once, and are transported from the starting workstation to the ending workstation in a linear, production-line like manner. This offers maximum throughput, with the rate-limiting step being the slowest workstation operation in the linear chain. For complex assays requiring numerous workstations, the cost of duplicating workstations makes these systems more costly than multitasking systems. However, as assays become less complex, requiring less workstation stages, this price gap narrows or may become nonexistent. The cost of linear-transport devices itself is less than general-purpose robotic arms. Linear systems that operate in this unidirectional manner do not require scheduling software.

Linear transport has been implemented in several ways. The most common is a derivation of true factory-style assembly line transport: a conveyor belt or moving track. The simplicity of conveyor transport systems makes them inherently a more reliable way to move plates among workstations. Workstations must be specifically designed or modified to accept a plate via this mechanism, in some cases with an active mechanism to "lift" the microplate off the track onto the workstation. Thus, these systems tend to be less adaptable to new devices due to the more limited nature of the transport system. Using small robotic arms to move plates to/from devices and the conveyor mechanism increases flexibility but reduces the speed and reliability inherent in a conveyor-based approach. System prices range from around $200,000 for small, focused "mini" systems to several millions of dollars for factory-scale systems.

Another linear approach utilizes multiple small, simple robotic arms to pass plates down the line, from workstation to workstation, not unlike a "bucket brigade." This transport mechanism combines some of the best features of both linear and multitasking systems. The use of a robotic arm at each workstation affords a high degree of flexibility in accessing each workstation, so adapta-

tion to new devices and technology is relatively easy. This approach also allows each workstation/arm unit to be treated as an independent module, which can be combined by the user with one or more other modules to form a variety of workcells based on the task at hand. However this flexibility is not without cost. Placing a capable robotic arm and controlling computer with each module makes this transport technique significantly more expensive than other linear approaches. The current example of this technique is the Zymark Allegro™ system, with prices ranging from $500,000 to over $1,000,000, based on the number of modules purchased.

2.6. Hybrid Approaches

There are several approaches that combine the features of both multitasking and linear transport. The first, currently offered by CRS Robotics Corp. (**Appendix 1**), uses a multitasking robotic arm to move stacks of plates to and from workstations ***(12)***. This technique places the workstations, not the robotic arm, in the rate-limiting role, similar to linear systems. Higher throughput can be achieved by simply adding more workstations. A limitation of this approach is that workstations must be capable of accepting and processing stacks of plates.

Linear-transport systems have also been configured to be bidirectional and multitasking to reduce the number of modules required and thereby reducing the cost of the system. In these cases, plates are moved both up and down stream with the linear transport and may re-visit a given workstation multiple times. These systems require scheduling capability just like multitasking robotic-arm transport systems, and differ from robot-based systems primarily in the mechanism of plate movement.

2.7. Detection

Detection is usually the final step of an HTS assay, which may or may not be integrated into an automated system. If an assay produces an end-product that is stable for a defined time, manually moving batches of completed assay plates to a detection device may be a very viable option. Ideally the detection instrument will accept stacks of plates, which is becoming more and more common. This approach simplifies the automated system, which is always desirable, and allows the detection instrument to be used for assays not connected to an automated HTS system.

If the final assay product is not stable, on-line detection is necessary. Most manufacturers of detection instruments today realize that their devices must easily interface with automation, and one can expect the new detection instruments to appear within automated systems fairly soon after introduction. Some detection devices incorporate liquid handling, reagent dispensing, and mixing

LUOs into their feature set, to facilitate very fast timing between final activities and detection. In multitasking systems, the timing of assay steps through detection can be maintained very precisely by the scheduling software. In linear systems, timing will be determined by the rate-limiting workstation, which in some cases may be the detection step.

2.8. Compound Library Storage, Retrieval, and Management

An HTS operation impacts both up and downstream company components. The number of compounds stored in corporate compound collections has grown along with screening capacity. Automation of the storage and processing of these collections becomes necessary to keep up with HTS efforts and minimize tracking and identification errors. The optimal choice of automation will be highly driven by the corporate organizational structure, i.e., whether the compound library and dispersal of samples is centralized or decentralized, whether the screening effort is centralized or decentralized and how geographically distant the various efforts are. In one extreme, the entire compound library may be stored adjacent to a central screening facility, both automated and linked by a plate-transport mechanism. In another extreme, compound storage may be automated only to the extent of bar-coded containers, and compounds are sent out to a variety of screening groups both nearby and around the world.

Technology originally developed for the automated storage and retrieval (ASRS) of industrial parts has been applied to the chemical collections. Some of the earliest examples were in the storage, inventory and processing of plutonium samples ***(13)***. Two basic types of ASRS have been employed for large centralized compound libraries. The first example stores multiple compound-containing drawers in a vertical rotary configuration, similar to a ferris wheel. The drawer with the desired compound (along with many others) is rotated to an access door, and extended for either manual or automated retrieval. These units range in height from 6–20 feet, and multiple units can be placed side by side to achieve storage capacity in the millions of compounds. Typically environmental control (humidity, temperature) is achieved by placing these units in an environmentally controlled room (usually custom built). The interface to further downstream automation or manual retrieval may be engineered as a "portal" to maintain environmental control.

The second example of industrial ASRS involves fixed storage racks or shelves, built linearly and to heights of approx 6–20 feet. A retrieval robot with vertical and reach axes of movement travels in front of the racks on a rail, moving samples from the racks to a fixed point at the end of the rail. These are much like automated fork lifts on a rail, having been developed for retrieving large bins or pallets in warehouses. Some models may access racks on both

sides of the rail. Environmental control must be achieved for the entire room housing the ASRS. Vendors of these systems include Aurora Biosciences, RTS Thurnall, and REMP.

These large systems, including the facility to house them will typically cost several millions of dollars. In both cases, storage of compounds may be done in bottles, vials, deepwell microplates, or regular microplates, in solid form, or dissolved in dimethyl sulfoxide (DMSO) ***(14)***. Solids are usually stored in a sealed vessel at room temperature with controlled humidity. The ideal mode of storing liquid compound libraries is still a topic of much debate, but HTS efforts are driven to work with DMSO-dissolved compounds due the speed of working with liquids vs the laborious nature of solids. Dissolved compounds are usually stored at refrigerated or freezer temperatures, often controlled humidity, and sometimes controlled inert atmosphere. However, below freezing environments and the formation of frost present a challenge for the mechanics of many automated devices. Automated devices for the handling and dissolution of solids do exist (The Automation Partnership), but are not universally applicable to a diverse collection of compounds.

Recently laboratory-scale ASRS devices have been developed. These store 100,000–250,000 compounds in fully automated, environmentally controlled carousels or elevators designed to hold either regular or deepwell microplates. These devices are specifically meant to fit into a normal-size laboratory. Plates are passed to/from the outside world through a portal, which may be integrated to typical plate transport and liquid handling automation. Vendors for these devices include Tecan, Zymark, and TomTec.

Compound library operations are often make large numbers of replicate plates for distribution to screening groups. Automated systems have been developed specifically for this operation, incorporating 96/384-channel pipetting workstations, extensive plate storage, bar-code reading, and applying capability, and plate-sealing technology. These generally fall into the Extended Workstation category described previously, and could be integrated with any of the available ASRS systems.

Any compound inventory system must be linked to a tracking database and often incorporates automated identification (**Subheadings 3.3.** and **3.4.**).

3. Integrating a System

System integration is the art and science of putting together electrical, mechanical, software, and communications technology to create an automated system that addresses the needs of the end-user, is efficient, reliable, and cost-effective. Fully integrated systems, both standard and custom, are available from a variety of commercial sources or one may purchase and/or build components and do the integration oneself if commercial systems are not appropri-

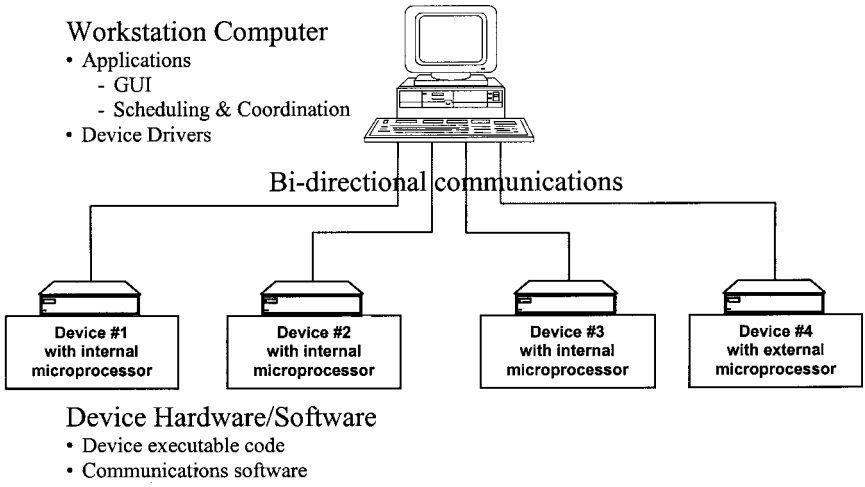


Fig. 1. System software architecture for a workstation. Adapted with permission from *(18)*.

ate. In either case, it is important to understand the fundamentals of system integration either to evaluate commercial systems or to plan and execute your own effort.

3.1. Software and Control

Well-structured automation systems incorporate the concepts of Computer Integrated Manufacturing (CIM) to define workcells and standard protocols to transmit information back and forth between the workcell(s) and the CIM-type software. This model scales to describe systems ranging from Simple Workstations to complex multi-workstation Integrated Systems. **Figure 1** shows this model applied to a workstation, where a central PC is interfaced to a number of devices such as a cartesian robot, syringe pump, peristaltic pump, water bath, and bar-code reader. These devices may have their own imbedded microprocessor and device code or can have this capability added by integrating an analog/digital interface board such as those available through National Instruments. The system PC contains GUI and system management software and drivers for each device. In essence, this is a small "integrated system."

Figure 2 shows this model scaled to a larger system, still controlled by a central computer housing the GUI, scheduling/management software, and device drivers. The only difference in this model is that the PC is interfaced to more complex workstations (i.e., a liquid handler, plate reader, or storage device), transport devices and a network environment. These naturally have more processing power on-board than would a syringe pump in the workstation model.

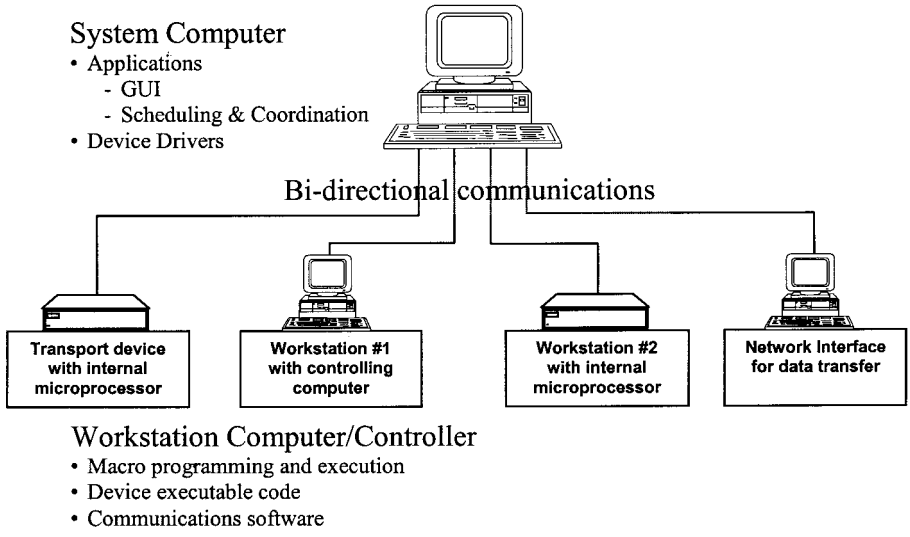


Fig. 2. System Software Architecture for an Integrated. System Adapted with permission from ***(18)***.

For example a liquid-handling workstation may have its own controlling PC and programming environment to create macro-level code that can be executed via the drivers on the system PC. But the basic model still remains intact.

The key to this model lies in the separation of the high-level application and driver code (on the system PC) from the lower level hardware, device, or workstation related software or firmware. This creates a modular environment where devices/workstations can be added (or taken out) with software changes only at the higher level, provided that necessary device driver code exists. Early efforts in laboratory automation often found device-related code at multiple levels, making modifications a very difficult and error-prone process.

The high-level system scheduling/management software may be as simple as code to prevent a liquid-handling cannula from colliding with a fixed wash station, or to assure that such a cannula is in liquid to prior to beginning an aspiration step. Such software is not usually apparent to the user. In more complex integrated systems, system scheduling takes on a higher profile and is offered as a specific product. Screening systems conduct multiple operations at several instruments or workstations concurrently, often requiring control over times for reactions, mixing, and incubation. Multitasking systems, i.e., those where the same workstation is used for several stages of the process, require interleaving of samples to maximize system throughput. While it is possible to

manually construct such an interleaving schedule, it quickly becomes a difficult Gantt chart exercise for all but simple operations. Software is now available to calculate an efficient interleaved schedule, and translate that schedule into executable code to drive the automated system. This software is nested into the GUI layer of a system and becomes a integral part of setting up and running the automation.

There are several distinguishing features of scheduling software. The most apparent is the style of the GUI. Text-based schedulers allow creation of an operation by choosing from a written list of potential operations or devices by choosing from a list or icon window. The created method "reads" textually like a written laboratory protocol. Graphical or icon-based schedulers offer the creation of an operation via drag and drop of icons or pictures, usually showing some visualization of the device or operation in question. The created method is a flow chart of icons or images, each of which can usually be opened for a verbal description of the operation behind the icon. The choice is a matter of user preference and has no impact on the eventual quality of scheduling. Most scheduling software offers a Gantt chart graphic depicting the completed schedule, estimated run times, either numeric or graphical representations of device percent utilization and postrun information such as error logs and a comparison of scheduled events to actual run-time events.

Scheduling engines can have different features and several levels of sophistication. Following are some of the important distinctions:

1. Linear scheduling: An algorithmic approach that puts the pieces of a schedule together starting with the first step for the first sample, moving step by step forward through the process for all subsequent samples. This is a fairly fast method, but usually does not create the most optimal schedule.
2. Hueristic scheduling: A "forward looking" mathematical approach that considers the entire list of steps and samples as one simultaneous calculation puzzle. Thus an operation at the end of the process, such as an incubation prior to reading can influence the way samples are spaced at the beginning, perhaps leading to a more efficient overall schedule. This calculation approach takes longer and/or more computing power.
3. Hard or variable event times: Some timed events in screening assays must exact, while others allow variance. Good scheduling software allows the user to define the level of variance allowed for each timing step. A window of variability in some steps can aid a heuristic scheduler in finding an optimal solution.
4. Single processes: The ability to schedule only one assay "thread," where only one plate moves from step to next step. Plates may enter or leave the process but multiple plates do not move from step to step. For example, a sample plate is transferred into an assay plate. The sample plate is then discarded or stored and does not continue to the next step while the assay plate continues.

5. Single, parallel processes: The ability to schedule only assay "thread," but allowing the creation of identical parallel tracks within that thread. For example, one sample plate gives rise to two duplicate assay plates, which then follow the same process.
6. Multiple processes: The ability to schedule different processes to run simultaneously or interleaved. For example, one sample plate gives rise to two duplicate assay plates which then follow *different* process.
7. Static schedules: A schedule that is calculated prior to the beginning of the run and whose timing is nonalterable during the course of the run. Because the scheduling is done prior to system operation, time-consuming algorithms can be used to achieve a highly refined schedule.
8. Dynamic schedules: A schedule that is calculated prior to the beginning of the run and whose timing or schedule is alterable or adaptable during the course of the run. The level of adaptability may range from a simple shift of timing due to a minor perturbation (i.e., an automatically recovered error) to complete recalculation of a schedule based on a sensed event (i.e., faster than expected timing, so speed up the schedule) or data input (i.e., based on the data from a plate read, branch to one of several next step options). Definitions of what "dynamic" means vary widely ***(16)***. Schedule recalculation on-the-fly must use fast, simpler scheduling algorithms and may therefore produce suboptimal schedules.
9. Simulation features: Many schedulers can execute scheduled code without the system devices actually receiving execution commands. This allows evaluation of the code performance short of actual mechanical movement. Some more sophisticated schedulers will generate a visual computer simulation of the system movement, and may report cases where the executed code would have resulted in a mechanical event such as a crash.

Scheduling software is offered by a number of system integrators (Appendix 1). Most schedulers today are not limited to interfacing with the devices and workstations supported by a specific system integrator. However, integrating one manufacturers scheduler with another's devices requires more in-depth knowledge of the software and driver architecture.

Communication between the system controller and workstations/devices may be accomplished in a variety of ways. The oldest style of interface is an analog signal, often a 4–20 ma current, which can, for instance, be used to control external motor speed. Simple on/off devices such as peristaltic pumps and valves may be controlled using contact closure or TTL (high/low) signals. More complex communications are done using serial, parallel or network pathways. As shown in **Table 1**, serial communications is still the predominant method of device communication, despite being an old technology. This includes RS-232, RS-485 and more recently USB (Universal Serial Bus) and HPSB (High Performance Serial Bus- aka IEEE1394, Firewire). Network connectiv-

Table 1
Percentage of Surveyed Using Different Types of Electronic Device Communication Modes[a]

	1997	1998	1999	Future
Serial	71%	74%	81%	54%
IEEE-488 (parallel)	48	46	53	51
4-20ma (analog)	52	39	37	23
Ethernet	37	38	43	40
USB 7	7	7	15	24
IEE-488HS	–	6	11	11
Other	20	25	38	39

a(Measurement Needs Tracking Survey—Keithley Instruments Inc. 1999)

ity such as Ethernet is growing in popularity, especially for the transmission of large sets of data from plate readers and imagers.

3.2. Error Handling

To take full advantage of the benefits of laboratory automation, unattended operation is often necessary. Such operation, however, requires that more attention be given to error avoidance, detection, correction, and reporting. Choosing a proper strategy must include sound system design, evaluation of sample stability, potential hazards, possibility of instrument damage, and the availability of trained personnel to correct problems ***(17)***.

A sound initial design is the best assurance of successful, error-free automation. The fact that additional error-handling capability is the most common system retrofit indicates that system design is often not given enough attention ***(18)***. First, equipment appropriate to the task must be chosen. Secondly, the chemical procedure may require modifications to compliment the strengths of the equipment and overcome the weaknesses. Finally, the system must minimize the possibility of automated or human error.

One can expect that errors will occur sometime during the operation of an automated system. The first step toward resolving error situations is detection of the error. Event sensing is one of the basic means of error detection. The appearance or nonappearance of an expected timed event such as data transmission can be a good check of the operation of a system. System schedulers can report and log discrepancies in expected timed events.

Screening systems generally involve moving liquids and objects, such as microplates, and it is desirable to verify that such movement did take place. Switches, optical sensors, and proximity sensors can be used to check opera-

tions such as proper valve rotation, the position of incubator doors, and correct placement of a plate prior to extending 96 liquid-handling cannulas into the wells of the plate. Many liquid-handling devices have liquid level-sensing capability, often capacitive, that can assure the cannula is below the air/liquid interface before aspirating. Robotic arms are equipped with some form of tactile feedback, which provides information about the force being exerted by the end-effector driving mechanism. This allows verification of the presence of a microplate or lid in the robotic grippers, and is probably the most widely used method of error detection in a screening system.

When an error is sensed, an appropriate action should be taken. The most desirable response is for the system to correct the problem and continue operation. Programming for error recovery must be sophisticated enough to account for variations in specific failures, including the possibility that the sensed failure is false. For instance, if failure to attach a pipet tip was sensed, an error-recovery routine must first execute a "discard tip" sequence before attempting once again to pick up a tip. Error-recovery routines should be programmed not lead to "error cascades," creating a larger problem than was initially present.

In some cases, automated error recovery may not be desirable, such as when precious compounds or reagents are involved. When an irrecoverable error is encountered, or recovery has been attempted and failed, the system may either bypass the error if this situation has been pre-determined to be acceptable or the system must be halted and the error condition annunciated in some way. Audible and visual alarms are effective where humans are likely to be nearby. Fully unattended systems can be programmed to send error notification through network or telephone connections to pagers, cell phones or e-mail. Systems may be switched back and forth between local and remote annunciation at different times of the day ***(19)***.

3.3. Automatic ID

Automated identification is essential in a modern high throughput laboratory. Bar coding has gained wide acceptance in screening labs as a specialized form of sample tracking and error avoidance. Bar coding is a visual representation of a digital code in which bars and intervening spaces represent ASCII (American Standard Code for Information Exchange) characters and symbols. Data transcription using bar codes has been estimated to be 1 billion times more reliable than manually entered data ***(20)***.

An identification strategy should be developed before any hardware is purchased. The fundamental choice is whether to use the bar code simply as a "pointer" to information in a database (also known as the "license plate" approach) or to imbed actual information into the bar code. The former strategy has positives such as simplicity and long-term scalability of scheme and the ability to use

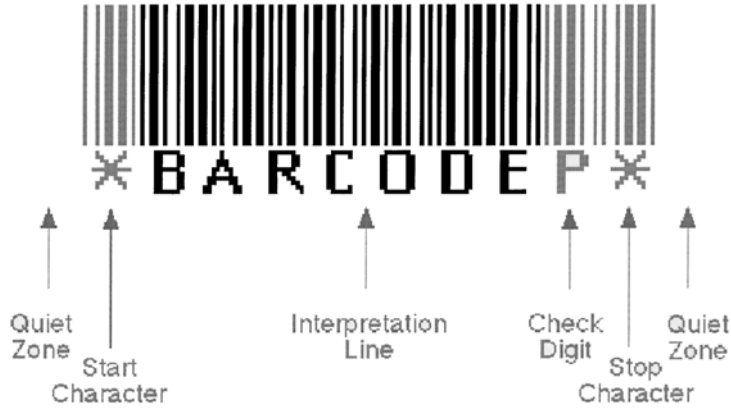


Fig. 3. General barcode structure. Adapted with permission from ***(18)***.

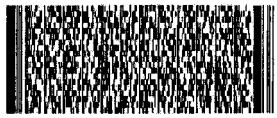
pre-printed labels of high quality, avoiding the cost of multiple in-house printing devices. Negatives of this strategy include reliance on regular and timely interaction with a database to transact ID data, and little or no human-interpretable information. The latter (imbedded information) strategy requires print-on-demand capability, often at multiple locations. Database interaction is less frequent, but still required to log or execute the creation of a new ID number. The most positive aspect of this strategy is that visual information on the label can identify the plate content without requiring database interaction. The largest downside of this strategy is the potential creation of an ID scheme that "evolves" over time with a tendency to imbed too much information into the label, thus becoming unscalable and unwieldy.

Bar codes are used in laboratories in two different popular formats. Code 3 of 9 (code 39) can express the full alphanumeric character set plus seven special characters (a total of 39). This code is among the least space-efficient and is therefore not always ideal for use on small labels such as microplates. Code 128 can express the full 128 character ASCII set, but more importantly is one of the least space-consuming codes, making it ideal for screening applications. Those considering new laboratory applications should evaluate Code 128 first ***(21)***. Both of these code types fit the general bar code structure illustrated in **Fig. 3**.

The demand for more detailed information in bar codes has led to the development of two-dimensional (2D) coding, often called dot or matrix codes. While not yet in the laboratory mainstream, they do represent the direction being taken in some industries, such as the Japanese Medical Association ***(22)***. Advantages of 2D code include much higher density of information and a high level of redundancy, which allows a significant portion of the label to be damaged yet still provide complete data. One version of 2D symbology is a stacked

500 characters, 10 mil linear—Code 39

**500 characters,
10 mil stacked linear—PDF 417**



**500 characters,
10 mil 2D matrix—Data Matrix ECC 200**

Fig. 4. Comparison of Bar Code Symbologies. Adapted with permission from ***(26)***.

bar code. These include the first 2D bar code, Code 49, introduced by Intermec Corporation in 1987. Since then additional formats have appeared, including PDF 417 and Code 1. Checkerboard or matrix symbologies can encode as many as several thousand characters in a small amount of space. This code consists of two black bars that intersect at right angles to define the perimeter of the code. Within this boundary, a series of black-and-white squares are written, as illustrated in **Fig. 4**.

Barcode readers are devices that will interpret the bar-code symbol and communicate the interpreted information to the laboratory automation system. The simplest of these devices use a laser or light-emitting diode to illuminate the code and use a light sensitive diode to receive the reflected pulses of light. These readers cannot decode a label that is smudged, torn, or incorrectly applied. More complex imaging devices, such as charged coupled device (CCD) cameras, can read the bar code in any orientation. These cameras have in the past been too expensive for routine use in automated systems, but dramatic price reductions now make them a more practical solution. Imaging scanners are required for many of the 2D code formats, such as Matrix.

The effectiveness of any bar coding strategy can by highly affected by the choice of label stock and the type of printer used. Label stock must be chosen after considering environmental conditions such as temperature, humidity, and chemical exposure. Synthetic stock rather than paper is more durable for a laboratory environment. If the label is to be placed in a freezer environment, the adhesive must remain viable at those temperatures, and also withstand freeze/thaw cycles. The adhesive must also be resistant to flowing around the

edges of the label, which can cause errors in automated microplate de-lidding and transport.

Externally printed labels, made by professional printing organizations, offer the highest quality of print resolution and durability. However, this requires an identification strategy that is compatible with pre-printed labels. In-house printing offers a much higher degree of flexibility. Numerous print technologies are available, including dot matrix, direct thermal, thermal transfer, ink jet, and laser printing. Dot-matrix printing will not offer adequate resolution to label most microplates. Direct thermal printing should be avoided since the paper is heat-sensitive. Thermal-transfer printing may be suitable in some cases, but does require a specialized printer as well as costly media and ribbons. A high-quality, single-pass, ribbon-type printer should be chosen to assure highest resolution printing. Laser printers will cost more than thermal transfer printers but do offer very high print resolution and low-cost consumables.

3.4. Data Transport and Management

Data is the ultimate "product" of screening groups, and automation has logarithmically increased the amounts of data being produced. It has also become more common for automated systems to require importation of key operational information. Therefore internal data management and bi-directional data-transfer capability are key features of screening automation (**Fig. 2**). The types of data handled by these systems include inventory, tracking, assay protocols, and measured data. Most commercial automation systems do not include a formal data-management package, but increasingly do provide tools to import and export data using industry standard protocols, such as ODBC compliant files, Excel format, and ASCII flat files. Similarly, most commercial screening data management systems do not offer plug-in data-management integrated with automated systems due to the complexity, specialization, and diversity of automation. This leaves a gap between the laboratory equipment and the database that must be filled by custom programming, often on a case-by-case basis, through either internal or contract resources ***(23)***. Some screening groups postpone dealing with this data-interface issue by using easily accessible software packages such as Excel. Inevitably these solutions lead to a crisis as increasingly large and complex data management requirements exceed the feature set of the "easy" tool. It is important to begin planning and executing a long-term data management and automation interface architecture long before the crush of data exists. A number of quality consulting groups have formed to fill the automation-data interface gap. These include Taratec Development Corporation (Bridgewater, NJ 08807); EMAX Solution Partners (Newtown Square, PA 19073); and Integrated Systems Consulting Group (ISCG) (Wayne, PA 19087).

3.5. Facilities

Anyone who has been involved in creating and growing a screening program has found that fully integrated screening systems are not made to fit into a typical wet chemical laboratory. Nor do most laboratories have the full range of services required by many of these systems. The ideal screening laboratory will have:

1. Access to utilities
   a. Compressed air (30 psi, 80 psi)
   b. Water (DI, chilled, heated)
   c. Vacuum (house, local)
   d. Gases
   e. HVAC (special ventilation)
   f. Power (conditioning, surge protection, UPS)
   g. Network
   h. Telephone (modem, pager, remote video)
2. Proper entry and exit for moving equipment
3. Space to walk completely around system
4. Compatibility with people, operation, and safety
   a. Isolate people from equipment
   b. Isolate equipment from people
   c. Isolate people from chemical/biological hazards
   d. Isolate equipment from hazards
      (Chemical, biological, radiological contamination, chemical spills, vapors)
   e. Provide for hazards detection
5. Storage space for disposables, waste and samples
6. Space for component test and repair
7. Preparation workspace

4. Future Direction

Current laboratory automation technology is poised to take the discipline forward for a number of years. The microplate format is almost universal, and all data indicates a deliberate movement toward higher-density microplates. The current transport technology is immediately compatible with higher-density formats, and liquid handling technology is progressing toward routine submicroliter capability. Systems built to handle 96-well microplates do not look greatly different from those designed for 384- or 1536-well microplates. It remains to be seen where practical limitations of hardware and biochemistry constrain the march toward microwell or micro-array approaches, but it is safe to say that microplates and automation to support that assay format will be around for some time to come.

4.1. Microfluidics

An emerging technology that has little lineage with current automation technology is microfluidics. Ironically, microfluidics has much in common with an older chemistry automation technology, flow injection, and segmented flow technology. The migration to nanoliter scale has been made possible largely through advances in micromachining driven by the semiconductor industry. Microfluidic systems are closed pumping systems, thereby having no evaporation problems, unlike microwell approaches. Detection must be sensitive and fast, but has the simpler imaging challenge of a serial sample flow rather than array imaging. The devices are or will be relatively simple, so the addition of parallel channels is not expected to be cost-prohibitive.

Significant challenges do still exist. The liquid handling interface of the macro world to the micro world is far from optimized, and currently tends to waste more sample and reagent than is actually used for the assay. Surface and fluid interactions have a significant effect since the very small flow channels (20–50 micron diameters) create a large surface-to-liquid ratio. One consequence is very strong laminar flow, which makes mixing of liquids difficult. Thermal effects, from the outside world or internally from chemical reactions have a large impact in the nanoworld. Pumping techniques, such as electroosmotic flow, can impact chemical reactions and mixing. The result is that many biochemical reactions behave differently in a microfluidic environment than in a microplate well. Screeners have spent years learning to understand and interpret biochemical and cell-based assay behavior in microplates. The same learning process will have to occur for the microfluidics environment. Early results indicate that the migration is possible, at least for certain classes of assays ***(24–25)***.

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Appendix 1: Technology Providers

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Aurora Biosciences Corp.
11010 Torreyana Road
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Fax: 858-404-6714
www.aurorabio.com

MDS Autolab Systems
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www.autolabsystems.com

BioDot Inc.
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Fax: 949-440-3694
www.biodot.com

BioRobotics Inc.
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Woburn, MA 01801
Tel: 781-376 9791
Fax:781-376 9792
www.biorobotics.com

Bohdan Automation
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Fax: 847-680-1199
www.bohdan.com

Caliper Technologies Corp.
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Fax: 650-623-0500
www.calipertech.com

Cartesian Technologies, Inc.
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Irvine, CA 92614
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Fax: 949-440-3694
www.cartesiantech.com

CCS-Packard
24030 Frampton Ave.
Harbor City, CA 90710
Tel: 905-332-2000
Fax: 905-332-1114
www.ccspackard.com

CRS Robotics Corp.
5344 John Lucas Drive
Burlington, Ontario
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Fax: 905-332-1114
www.crsrobotics.com

Cyberlab, Inc.
36 Del Mar Drive
Brookfield, Ct. 06804
Tel: 203-740-3565
Fax: 203-740-3566

CyBio, Inc.
500 West Cummings Park, Suite 1200
Woburn, MA 01801 USA
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Fax: 781-376-9897
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www.cyber-lab.com

EMAX Solution Partners
18 Campus Blvd., Newtown Square
Pennsylvania, 19073
Tel: 610-325-3700
Fax: 610-325-3782
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Hudson Control Group
44 Commerce Street
Springfield, NJ 07081
Tel: 201-376-7400
Fax: 201-376-8265
www.hudsoncontrol.com

Intelligent Automation, Inc.
149 Sidney Street
Cambridge, MA 02139
Tel: 617-354-3830 (ext. 401)
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www.automationonline.com

Orchid Biocomputer Inc.
303 College Road East
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www.orchidbio.com

Motoman, Inc.
805 Liberty Lane
West Carrollton, Ohio 45449
Tel: 937-847-3300
www.motoman.com

Oyster Bay Pumpworks
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Fax: 516-624-9253
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Rixan Associates
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Source for Automation
327 Fiske Street
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Tecan U.S.
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TomTec
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