Microdispensing technologies in drug discovery

Don Rose

Because of the advent of managed care,the pharmaceutical industry is entering a new era, characterized by increased competition and pricing pressures. As a result, drug discovery within pharmaceutical companies is rapidly embracing new paradigms to help bring more novel drugs to the market as rapidly as possible. One paradigm currently being pursued is the miniaturization of the processes involved in the exploratory phase of drug discovery. This reduction in scale has led to the development of new dispensing technologies. This review examines several microdispensing technologies for drug discovery.

Drug discovery is entering a new era and pharmaceutical companies are experiencing increasing market pressures to deliver novel drugs in a timely manner that address unmet medical needs. At the same time, new technologies are being developed that will greatly accelerate the drug discovery process. The areas of intense technological change include compound synthesis, genomics and drug screening. Compound synthesis has undergone a major paradigm shift with the advent of combinatorial and parallel synthesis, a technique for generating thousands of compounds per synthesis cycle. Genomics research, spearheaded by the Human Genome Project and its related technological advances, has greatly expanded the number of novel drug targets. As a result of the increased numbers of targets and compounds, drug screening has begun to incorporate automation that was previously found in analytical chemistry laboratories, or on manufacturing floors, to create high-throughput screening (HTS).

One common theme among many of these technological advances has been a reduction in the scale of these techniques. Whether a chemical synthesis, biochemical assay or polymerase chain reaction (PCR), the scale is steadily being reduced in an effort to cut reagent costs and increase sample throughput. This reduction in scale has created a need for microscale liquid handling – the dispensing and transfer of submicroliter volumes. This article reviews the emerging technologies that are useful for microdispensing as they apply to drug discovery. It should be noted that, because of the recent emergence of this area and the fact that the field is developing primarily outside academic areas, this review contains little published literature and is based primarily on trade journal descriptions, product literature, conference proceedings and customer applications.

Microdispensing technologies

In drug discovery, most automated liquid handling is accomplished using either syringe pump-based pipettors for transferring fluid, or peristaltic pumps for reagent filling. Syringe pump pipettors use a syringe coupled to a metal or plastic tip. Displacement by the syringe in turn displaces fluid from the tip. Typically, these pipettors can dispense over the range of low microliters to milliliters.

As an interim solution to provide submicroliter dispensing, syringe pump pipettors can be configured to produce a small drop on the tip that can be touched to a dry surface for dispensing (touch-off dispensing). However, to provide more versatile dispensing and to reduce drop size

Don Rose, Cartesian Technologies, Inc., 1906 E. Hwy 54, Suite 200A, Durham, NC 27514, USA. tel: +1 919 572 6888, fax: +1 919 572 6889, e-mail: drose@cartesiantech.com

 1359-6446/99/$ – see front matter PII: S1359-6446(99)01388-4

to the nanoliter or picoliter dispensing range, new technologies are required. Two technologies that do dispense in the nanoliter and picoliter range are inkjet and pin-transfer technologies.

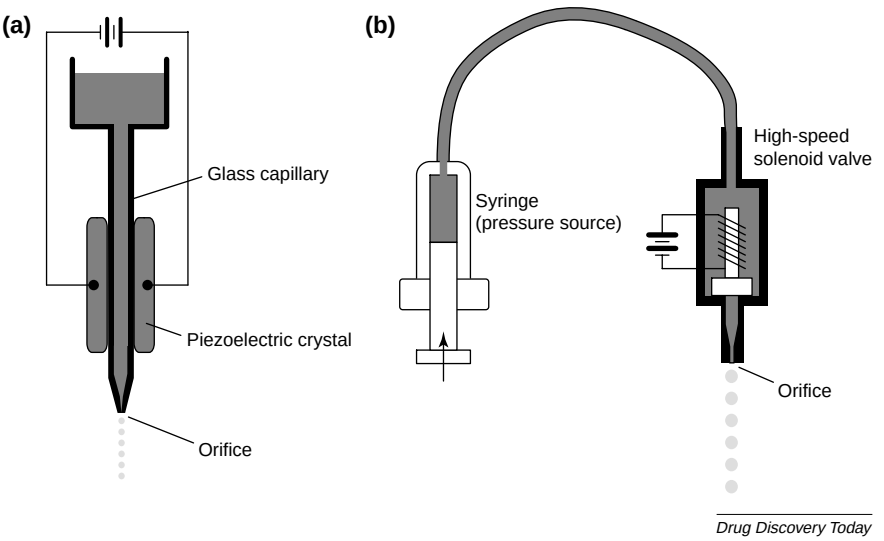

Figure 1. *Schematic of inkjet dispensing technologies: (a) piezoelectric; (b) syringe–solenoid. Piezoelectric dispensing is accomplished by squeezing a glass capillary with a piezoelectric crystal to force liquid through an orifice and eject a drop. Syringe–solenoid dispensing is accomplished by creating a positive pressure with the syringe pump and opening the valve for several microseconds to release the pressure and eject a drop.*

Inkjet microdispensing technologies

Inkjet microdispensing is derived from the inkjet printing industry. The basic principle of inkjet microdispensing involves the application of force to create a rapidly moving liquid stream, which then passes through a small orifice. On passing through the orifice, the stream achieves sufficient linear velocity to overcome surface tension forces and is ejected in the form of a drop.

Inkjet dispensing technologies have several advantages over conventional syringe pump-based dispensers. First, they can create small drops that range in volume from nanoliter to picoliter. In addition, because the technique is non-contact, delivery of the drop to a small location is not limited by the mechanical size of the tip. For example, a drop can be delivered into a 2 mm-diameter well simply by ejecting the drop above the well without the tip entering the well. Finally, the speed of inkjet printing translates into high-speed dispensing when using these technologies.

The two most common types of inkjet dispensing technologies are shown in Fig. 1. Piezoelectric dispensing (Fig. 1a) uses a piezoelectric crystal that is in contact with a glass capillary tube to apply force to the fluid. Applying a voltage across the crystal causes it to deform the wall of the capillary with the subsequent ejection of fluid from the tip of the capillary. Typical piezoelectric dispensers can create drops in the picoliter range and can operate at rates of up to 1000–5000 drops per second. However, given the small size of the orifice and its dependence on surface tension, piezoelectric dispensing can be prone to clogging (e.g. by compounds precipitated in dimethylsulfoxide, DMSO) and intermittent dispensing caused by gas bubbles.

Another type of inkjet dispensing technology couples the high-resolution displacement capabilities of a stepper motor-driven syringe pump with the high-speed actuation of a microsolenoid valve. This syringe–solenoid technology (Fig. 1b) uses the syringe pump to create a steady-state hydraulic pressure upstream from the microsolenoid valve. Upon actuation of the valve, a rapidly moving stream of liquid moves to the orifice and is ejected as a drop. The technology can be used in two different modes, as shown in Fig. 2. Using the syringe pump, the sample can be aspirated from a source and then dispensed into a destination location (Fig. 2a). Alternatively, the sample can be introduced through the entire fluidic path and dispensed in a continuous fashion (Fig. 2b).

This patented technology[1,2] can dispense discrete drops over a wide range of volumes in the low nanoliter to low microliter range, as well as up to hundreds of microliters as a stream of fluid. Because of the positive displacement nature of the dispense mechanism, air bubbles and fluid surface tension have only minimal effects on dispense precision and accuracy. Typical dispense accuracy for this technology is shown by a standard error (SE) of 2–3% and the reproducibility by a coefficient of variation (CV) of 3–10%, depending on the volume.

Pin-transfer technologies

Pin-transfer technologies involve the use of a rigid pin to transfer liquid from a source to a destination. The process involves dipping the pin into a sample, withdrawing the pin, which results in a small volume of liquid on the tip of the pin, and then placing the pin on a solid surface to

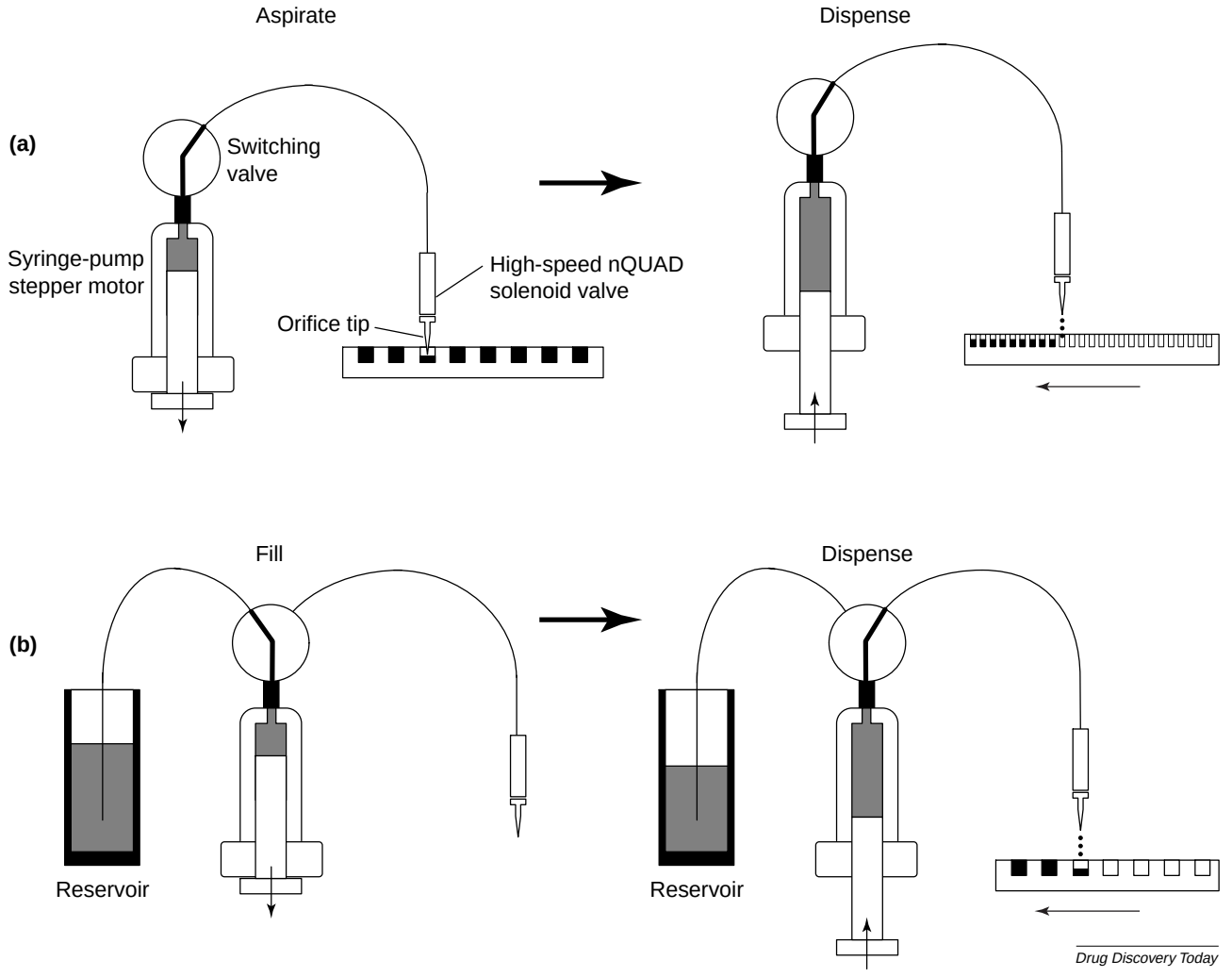

Figure 2. *Two modes of syringe–solenoid inkjet dispensing: (a) aspirate–dispense; (b) bulk dispense. Aspirate–dispense involves dipping the orifice tip into the source sample, withdrawing the syringe to bring sample into the tip and moving the tip to a destination position for dispensing in a non-contact manner. Bulk-dispense involves filling the entire fluidic path with reagent and then dispensing to a series of destinations.*

deposit the volume. Pin transfers were first done using solid pins (Fig. 3a), the tips of which were ground to a specified point depending on the volume to be transferred, typically low nanoliters. Other pin variations have been developed more recently to permit more than one transfer per dip into the sample. For example, split or quill pins (Fig. 3b), which were pioneered at Stanford University, capture the sample in a slot such that touching the pin onto a solid surface results in the deposition of a small spot, typically in the mid-picoliter volume range.

Pins have the advantage of being simple, robust, relatively inexpensive and small enough to arrange in a high-density array. However, compared with inkjet dispensing, the spot-to-spot variation can be much higher, in the order of 10–25% CV, because of variations in pin geometry and surface chemistry. Furthermore, pins dispense a small but fixed volume, which can limit their utility for applications requiring variable volumes.

Application of microdispensing technologies to drug discovery

Practical issues

The application of microdispensing technologies in drug discovery requires a thorough understanding of the variables

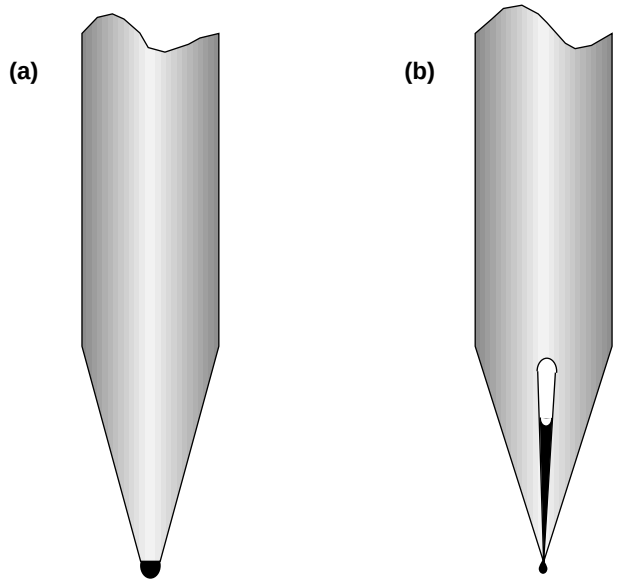

Figure 3. *Schematic of different types of pins for contact printing: (a) solid pin; (b) split or quill pin. The solid pin holds a small volume of sample on its tip, whereas the quill pin holds the sample within a slot, allowing multiple spottings per loading.*

that can affect liquid handling. Many of the variables that are not important with conventional liquid handling become critical as the dispense volume decreases.

Solution properties. As the size of the dispensing volume decreases, the properties of the dispensed liquid will have a greater effect on the dispensing mechanisms. For example, surface energy or tension of the liquid has a dramatic effect on how a drop breaks away from the orifice of an inkjet-type dispenser. For pin printing, many more surface properties come into play. For example, when a pin touches a glass surface, the interaction between the surface energies of the pin, the glass surface and the liquid determines the quantity of liquid deposited onto the surface.

In general, the more viscous the sample, the more difficult it is to dispense because of difficulties in drop breakaway, be it from a pin or an inkjet orifice. The dispensing of suspensions (e.g. cells) can be done using either of these technologies. However, if the particles are large relative to the orifice or the gap in the pin, clogging can occur. Cells have been dispensed using both piezoelectric and syringe–solenoid technologies with very little damage to the cells.

Static charge. As drops become smaller, they can be affected by static charge. This is seen predominantly with inkjet-type dispensers where drops are deposited onto a surface. For example, drops of a DNA solution, which by virtue of the phosphodiester backbone can acquire a net negative charge, will have an altered trajectory when dispensed onto a surface with a net positive charge. This problem can be solved by flooding the surface with ions from an ionizer.

Evaporation. As the volume of a liquid decreases, the surface area–volume ratio increases dramatically, which translates into a higher rate of evaporation. Evaporation of drops during transit from an inkjet dispenser is usually not an issue except for drops of low picoliter volume. However, evaporation becomes more of an issue for liquids after dispensing, as well as for liquid in source plates for high-throughput systems that are processing a number of plates over an extended period of time. This evaporation can be reduced by using a humidified chamber.

Mixing. As the size of the volume is reduced, mixing must be achieved using different modes. For example, with a 96-well plate, which can hold several hundred microliters of liquid per well, mixing can be accomplished by shaking the plate. However, shaking is ineffective for a 1536-well plate with only a few microliters of liquid per well, because of the surface tension of the liquid in each well.

Mixing on a small scale can be achieved using the dispensing mechanism. For example, the linear velocity of a series of drops emanating from an inkjet dispenser can cause mixing of reagents in a small well. Furthermore, diffusion can become a dominant means of mixing volumes of only a few microliters.

Figures of merit. In evaluating and comparing new technologies, several figures of merit are important to understand:

- Linearity is a measure of the degree to which changes in the predicted volume correspond to changes in the actual volume. For example, doubling the programmed volume from 100 nl to 200 nl should result in double the volume (or a read-out signal that correlates to the volume). In an ideal case, a plot of expected volume versus actual volume will result in a correlation coefficient (R^2) of 1.00. In reality, R^2 values of between 0.9900 and 0.9999 are acceptable.
- The dynamic range is the volume range over which the dispenser is linear. For some applications, dynamic range is important as it determines the flexibility of a dispensing system. For example, a dispensing technology that covers a dynamic range of 10 μl will have less utility compared to one that covers a dynamic range of 250 μl.

- Precision, often referred to as reproducibility, is the extent to which a group of measurements differ from one another. Precision is determined by measuring the mean and standard deviation of a set of volumes (usually greater than ten measurements). A common method for stating precision is in terms of the relative standard deviation (RSD) or CV and is calculated as follows:

  RSD or CV (%) = 100 × (SD ÷ mean)

For drug discovery applications, this figure of merit is often the most important figure and must be below 10% CV, and preferably 5% CV for some applications.

- Accuracy is a measure of the variation of the actual drop volume from the expected drop volume and is calculated as a standard error (SE):

  SE = 100 × [(actual volume − predicted volume) ÷ (predicted volume)]

A standard error of less than ±5% is usually acceptable for most applications.

- Throughput is a measure of the number of dispenses per unit time and must be normalized for each application. For example, although most inkjet-type dispensing is very fast, its speed is only realized for multiple dispenses of the same reagent. If washing is required between each transfer, then the speed is similar to that of conventional dispensers.
- Robustness or reliability is a qualitative measure of the long-term performance of the technology as implemented in an instrument. In other words, how does the technology perform with repeated daily use, which parts fail and which need to be replaced on a routine basis.

As an illustration of the use of figures of merit, test dye was aspirated and dispensed at different volumes and measured using a plate reader. The correlation between the actual and the predicted dispense volume is shown in Fig. 4. Table 1 shows the reproducibility of the dispenses.

HTS microdispensing applications

In HTS, there has been a move from the conventional 96-well plates to the utilization of higher density plates containing 384-, 1536-, 3456- and 9600-well plates. This move is being driven by a need to reduce reagent costs as well as to obtain more information from each plate run through the screening process. For example, a cost–benefit analysis carried out by a major pharmaceutical company compared the cost of screening singlet compounds in 96-well plates to that of triplicate screening in 9600-well plates. The details of the analysis are shown in Table 2.

Compound transfer. The first step in the HTS process is the creation of assay plates by transferring the compound (typically compound dissolved in a suitable solvent such as DMSO), from a master plate to the assay plate. The quantity transferred needs to be small enough such that the DMSO concentration in the final assay volume is less than 1–2%. Because of the limitations of conventional dispensers in delivering small volumes, compounds are typically transferred to an intermediate plate, diluted and then an aliquot is transferred to the assay plate, posing several limitations: the dilution step decreases the storage life of the assay plate (i.e. aqueous hydrolysis of the compound can occur), and it wastes compound.

Table 1. Reproducibility of the dispenses

Dispense volume (nl)	CV (%)[a]
8.33	10.22
20.8	9.77
41.7	5.70
70.8	5.44
100	3.74
250	3.22
500	6.70
1000	4.54
5000	2.45
10,000	1.98

[a]Each value represents the coefficient of variation (CV) from 96 (12 by 8 channels) dispenses.

Table 2. Comparative costs (in US$) of screening compounds singly or in triplicate

	96-well plate (single screening)	9600-well plate (triplicate screening)
Plate costs	4000	200
Peptide reagent costs	25,000	150
Enzyme production costs	150,000	50,000
Labor costs	30,000	2500
Total costs[a]	209,000	53,000
Savings for one assay per year	–	156,000
Savings for 32 assays per year	–	5,000,000

[a]Based on a 100,000-member compound library.

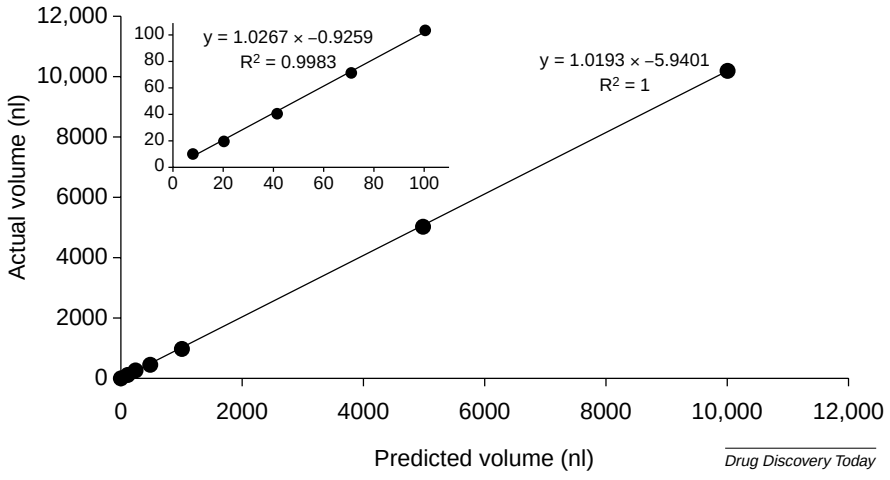

Figure 4. *Data showing linearity, accuracy and dynamic range for syringe–solenoid dispensing technology. The inset shows linearity at low volume. A test dye (tartrazine in water) was aspirated and dispensed using an eight-channel syringe–solenoid-type dispenser (PixSys 3200, Cartesian Technologies, Irvine, CA, USA). Each data point represents the mean for 96 (8 by 12) dispenses.*

Microdispensing technologies can solve this problem by directly transferring the compound from the master plate to the assay plate, thereby eliminating the intermediate dilution step, permiting 'just-in-time' assay plate preparation. For example, the largest volume containing DMSO-dissolved compound that can be transferred from a 96-well plate to another plate type, while keeping the DMSO concentration at 1% in the final assay volume, is 2 μl for a 96-well plate, 0.5 μl for a 384-well plate, 100 nl for a 1536-well Greiner plate and 20 nl for a 1536-well Corning plate. Thus, as one moves to the higher density (lower volume) plates, nanoliter microdispensing is necessary to keep the DMSO concentration below 1% without intermediate dilution steps. Most conventional dispensers have difficulty in reliably dispensing volumes below 1 μl, which limits the use of 384-well plates. Furthermore, with the non-contact technologies such as syringe–solenoid and piezoelectric, just-in-time dose–response curves can only be created over a limited range without dilution.

Conventional syringe pump dispensers have been modified to transfer as little as 200 nl with touch-off in a 1536-well plate[3]. Syringe–solenoid technology has been used to transfer 20–100 nl of dissolved compound from 96-well plates to 9600-well plates[4], whilst piezoelectric technology has been shown to transfer compound over a 128-fold volume range (0.28–36 nl) to produce an IC_{50} dilution series[5].

Although not widely used, pins offer a possible alternative to non-contact dispensing if throughput and cost are more important parameters than precision, accuracy and programmable dispense volume. To use pins for compound transfer, the pin is dipped into the sample well then spotted onto the bottom of a dry microwell plate. A pin dispense head could be configured with 96 pins, making the transfer process very rapid.

Reagent dispensing. After assay plates have been prepared, the assay is conducted by a series of reagent additions and incubations followed by a read-out of the results. As noted above, because screening operations are moving to the use of higher density plates, rapid, low-volume reagent dispensing is required. The typical dispensing volumes span a wide range (typically 10–80% of the well volume): 2–50 μl for a 96-well plate, 0.5–30 μl for a 384-well plate and 0.02–8 μl for a 1536-well plate.

Syringe-pump dispensing can be used for reagent addition to high-density plates, but because touch-off dispensing is difficult (reagent or compound usually exists in the well prior to dispensing), addition volumes are limited to several microliters[3].

Reagent addition using syringe–solenoid inkjet technology can be accomplished in one of two ways, as shown in Fig. 2. The reagent can be aspirated from a common reservoir and dispensed across the plate (Fig. 2a), or the reagent can be plumbed through the system and dispensed in a continuous mode across the plate (Fig. 2b). The advantages of the aspirate–dispense mode are less reagent loss and the ability to transfer several different reagents, whereas the bulk-dispensing mode is much quicker. Dispensing several reagents simultaneously can be accomplished by using a multichannel dispenser and either aspirating samples from different reservoirs or plumbing different reagents through each channel from different reservoirs. In either case, separate valves can be actuated to dispense each reagent individually.

Dispensing into the high-density plates using the syringe–solenoid technology can be carried out by moving the dispensing head over the well, dispensing the drop into the well, and then moving to the next well. A faster

method involves coordinating the ejection of drops from the inkjet valve with the movement of the table, as shown in Fig. 5. Here the actuation frequency and the table speed are adjusted to deliver the correct number of drops to each well in a rapid manner. With this type of 'on-the-fly' dispensing, a high-density plate can be filled in tens of seconds, whilst Oldenburg and coworkers were able to fill a 9600-well plate with 200 nl in 90 s using a four-channel system[4].

Genomics applications

Microarrays. These microarrays are high-density arrays of genomic material (synthetic oligonucleotides or cDNA clones) placed on a solid surface, each individual element representing a unique sequence of DNA. When a sample, labeled with a fluorescent tag, is applied to the array, those sequences that are complementary to the sequences in the array will hybridize. Washing away the excess sample and reading the array reveals the identity of the sequences in the sample.

Genomic microarrays were first pioneered by researchers at Affymetrix (Santa Clara, CA, USA where the arrays were made by photolithographic synthesis of oligonucleotides on a glass substrate[6–8]. More recently, alternative methods of creating microarrays have subsequently been developed, the most popular being the use of pin-transfer technology to spot DNA samples onto glass microscope slides[9–13]. This application uses a split- or quill-type of pin, which when dipped into a well containing an oligonucleotide sample, can be spotted onto multiple glass slides. An example of this type of spotting is shown in Fig. 6. The center-to-center distance of the spots is 300 μm and the spot diameter is approximately 150 μm. Spacing in this range will permit approximately 5000–30,000 spots to be printed onto a slide.

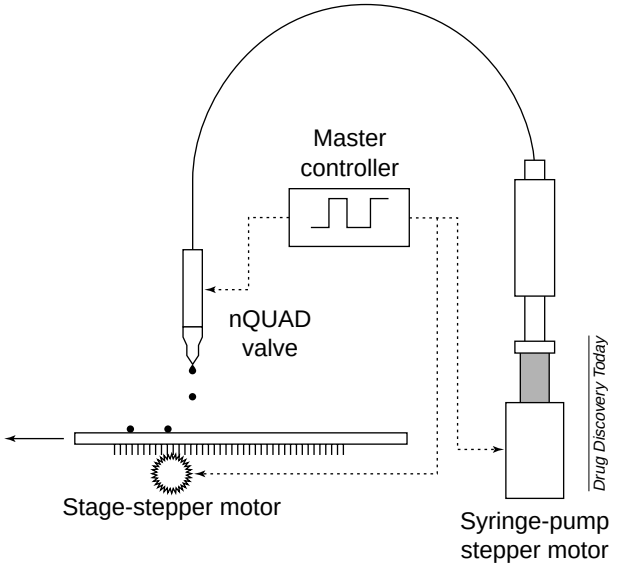

Figure 5. *Synchronized dispensing of drops using syringe–solenoid dispensing. This configuration permits 'on-the-fly' dispensing, much like a desktop printer (patent pending, Cartesian Technologies).*

Although experimentally different (the actual experiment spots non-fluorescent DNA and hybridizes with fluorescently labeled DNA), spotting fluorescently labeled DNA gives some indication of spotting reproducibility. Table 3 shows the reproducibility of pin-spotting for the three numbered arrays in Fig. 6.

Spot-to-spot variation results primarily from changes in properties of the glass surface over the slide, optical variations during the scanning process and changes in the volume held within the pin during the spotting process. The spot-to-spot variation will increase significantly when multiple pins, multiple slides and variations in slide-attachment chemistry are introduced.

An alternative pin-technology for creating microarrays uses a 'pin-ring'. A small ring is dipped into the sample to withdraw a small volume, which is held in the ring by surface tension. A solid pin is lowered through the ring to place a small volume of sample on the end of the pin. Further lowering of the pin spots the sample onto the surface[14].

Syringe–solenoid inkjet dispensing has been used for printing arrays, but the spot densities are typically between 200 and 400 spots per square centimeter. Smaller spot sizes can be achieved with piezoelectric inkjet dispensing. For example, spots of diamter 165 μm (325 pl), which are suitable for genomic arrays, have been generated (Scott Levy, Packard Instrument Company, Meriden, CT, USA, pers. commun.). A novel approach to creating microarrays involves piezoelectric inkjet dispensing of oligonucleotides in a line pattern on a membrane. The membrane is rolled up 'jelly-roll' style and sliced to produce circular arrays[15].

Low-volume reactions. Another application area for microdispensing in genomics involves low-volume reactions. For groups carrying out a large number of sequencing reactions or PCR reactions, the reagent costs become significant. Reducing the volume of these reactions not only increases throughput but also reduces the costs of the reaction. As with HTS, a reduction in volume can only be accomplished with the technology and equipment to dispense small volumes.

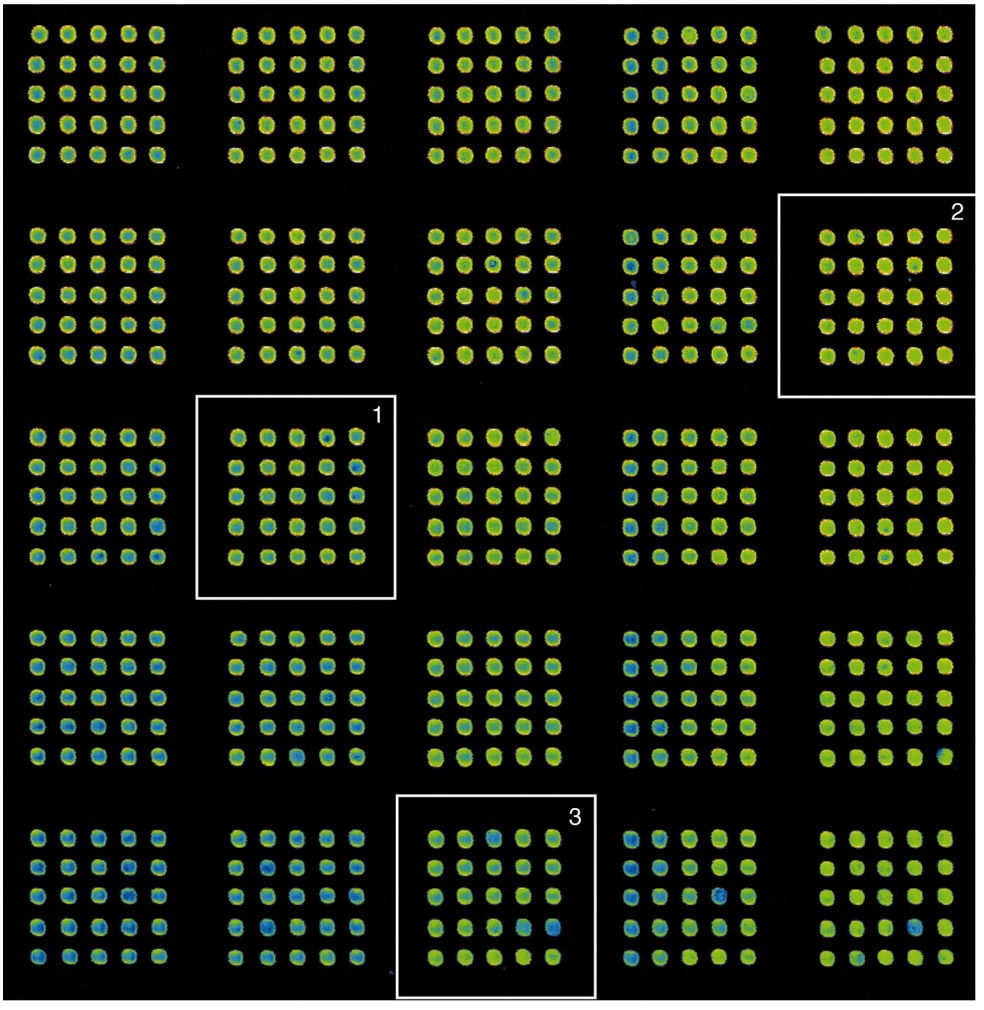

Figure 6. *Fluorescently labeled 30-mer spotted onto a glass slide using a single quill-type pin. Center-to-center spot distance is 300 μm and spot diameter is approximately 150 μm. The array was printed by dipping into the sample, printing five 5 × 5 arrays in column 1, washing, and repeating the printing for the subsequent columns. Details of the reproducibility of the arrays numbered 1–3 are given in Table 3.*

making microscale dispensing more useful for chemical synthesis. Finally, the nature of the solvents and reagents used for chemical synthesis places constraints on microdispensing technologies, not found in other areas where primarily aqueous solvents are used.

Currently, a typical microscale chemical synthesis involves several hundred milligrams of resin in a 96-well plate format. The dispensing volumes for these formats are in the high microliters, not requiring the use of any microdispensing technologies. If synthesis moves to 384- or 1536-well plate formats, then microdispensing technologies would be more applicable.

One example of combinatorial synthesis using microscale dispensing[16] used a pneumatic-solenoid inkjet technology to dispense to a novel format for synthesis. The dispenser, the ChemJet, is composed of 48 individual channels and is capable of dispensing 1–8 μl, each of 48 reagents onto a molded polypropylene sheet consisting of a 48 by 48 matrix of 8 μl-wells (the ChemSheet). The sheet was chemically grafted to allow solid-phase synthesis in each well and dispensing was carried out in an orthogonal manner (right to left, then top to bottom) to provide a combinatorial mix of reagents.

Combinatorial chemistry applications

Library synthesis. For several reasons, the requirement for microdispensing technologies in combinatorial- or parallel-library synthesis has lagged behind that in HTS and genomics. Primarily, the effort to reduce the scale of chemical synthesis has not reached the same level as in the other areas, perhaps because chemical synthesis has traditionally been carried out on a large scale (liters and grams). Furthermore, the relatively large quantity of compound required for analytical characterization and biological assays has limited the movement of synthesis to smaller scales. As better analytical methods are developed and the scale of assays decreases, the quantity of compound required will decrease,

Bead dispensing and handling. Most combinatorial synthesis uses solid-phase reactions on derivatized beads or resin. As the scale of chemical synthesis decreases, the dispensing and handling of these beads becomes critical and requires some microscale techniques. Currently, resins are usually distributed into 96-well plates by pumping a slurry of beads through a multichannel dispense-head using a peristaltic pump. However, after synthesis or assay, there is often a need to handle very small numbers of beads. For example, to sample 10–30 beads from a large batch, a conventional

Table 3. Reproducibility of pin-spotting

5 × 5 array[a]	Mean intensity	Standard deviation	CV (%)
1	23,363	1926	8.2
2	31,252	2451	7.8
3	19,186	2439	12.7

[a]Arrays are shown in Fig. 6.
Abbreviation: CV, coefficient of variation.

syringe pump can be used to aspirate an aliquot of the beads. Handling single beads requires much more sophisticated technology. Recently, a technique has been developed to pick single beads from a sample and dispense them into a discrete location (J.C. Nelson *et al.*, GlaxoWellcome, Research Triangle Park, NC, USA, unpublished). The technique uses conductivity detection to sense the presence of a bead on or at the probe tip, and optical detection to determine the size of the bead (single, double or fragment).

Summary and future directions

Microdispensing technologies will play a more important role in drug discovery programs as the fields of HTS, genomics and combinatorial chemistry mature. The need for microdispensing technologies in HTS will be driven by the following:

- *Reagents and formats.* Fluorescent reagents and high-density formats for carrying out assays on a microvolume scale must be developed further. For example, the ability to carry out scintillation proximity assays in a 1536-well plate will greatly accelerate work in higher density plates. Furthermore, the development of a 384- or 1536-well filter-plate would make many analyses more amenable to high-density assays.
- *Instrumentation.* Although syringe–solenoid technology is commercially available (Cartesian), piezoelectric dispensing for HTS is still under development (Packard). Likewise, very few detectors are available for high-sensitivity detection of the higher density plates (LJL BioSystems, Amersham, UK). As the instrumentation becomes more widely available, movement to higher density formats will increase.

The future role of microdispensing technologies in genomic microarray applications differs relative to HTS applications. Microarray applications are very new and are not undergoing the same evolutionary migration as HTS (i.e. 96- to 384- to 1536-well plates). The need for microdispensing technologies in microarrays will be driven by the following:

- *Application development.* As the application areas for microarrays become wider, there will be a greater need for microdispensing, as well as for detection technologies and instrumentation. For example, microarrays are currently being used for gene expression profiling and genotyping in model organisms such as yeast. In the future, applications will broaden to include human and plant tissue in toxicological, pharmacological and clinical applications.
- *Patents.* A number of key patents are held in the microarray field and the manner in which these patents are enforced will determine the need for microdispensing technologies.

The future role of microdispensing technologies in combinatorial chemistry will follow the path of HTS: movement from 96- to 384- to 1536-well plates. The rate of this migration will be slower than for HTS and will depend on the downstream requirements of compounds for analytical characterization and biological assays, as well as the availability of automated instrumentation to perform chemical reactions on a microliter scale.

REFERENCES

1 Biodot (Tisone, T.) (1998) US Patent 5741554
2 Biodot (Tisone, T.) (1998) US Patent 5738728
3 Comley, J.C.W., Reeves, T. and Robinson, P. (1998) *J. Biomol. Screening* 3, 217–225
4 Oldenburg, K.R. *et al.* (1998) *J. Biomol. Screening* 3, 55–62
5 Driscoll, J. *et al.* (1998) *J. Biomol. Screening* 3, 237–239
6 Kozal, M.J. *et al.* (1996) *Nat. Med.* 2, 753–759
7 Chee, M. *et al.* (1996) *Science* 274, 610–614
8 Wodicka, L. *et al.* (1997) *Nat. Biotechnol.* 15, 1359–1367
9 Shalon, D., Smith, S.J. and Brown, P.O. (1996) *Genome Res.* 6, 639–645
10 Schena, M. *et al.* (1995) *Science* 270, 467–470
11 Cheung, V.G. *et al.* (1998) *Nat. Genet.* 18, 225–230
12 Heller, R.A. *et al.* (1997) *Proc. Natl. Acad. Sci. U. S. A.* 94, 2150–2155
13 Solinas-Toldo, S. *et al.* (1997) *Genes Chromosomes Cancer* 20, 399–407
14 Rose, S.D. (1998) *J. Assoc. Lab. Autom.* 3, 53–56
15 Stimpson, D.I. *et al.* (1998) *BioTechniques* 25, 886–890
16 Lemmo, A.V. *et al.* (1997) *Anal. Chem.* 69, 543–551