Automated compound management evolves to meet the drug discovery demands of the future

The development of new drugs involves testing hundreds of thousands of compounds, from high throughput screening through to subsequent lead optimisation. Recognising the importance of a compound bank in supporting drug discovery activities, as well as to accommodate the change from standardised HTS to integrated lead finding, the Compound Management group at the Novartis Institutes for Biomedical Research embarked on an innovative compound bank initiative to meet the demands of today's research – the implementation of an advanced automated tube store and dose-response plate module.

By Dr Caroline Engeloch and Dr Daniel Baeschlin

The Novartis Institutes for Biomedical Research (NIBR) is dedicated to developing groundbreaking treatments and cures for currently unmet medical needs, testing hundreds of thousands of chemical compounds at its sites in Cambridge, USA and Basel, Switzerland, for lead optimisation. High throughput screening (HTS) plays a key role in lead discovery – the identification of active compounds which, individually or in combination, may ultimately become an effective drug. Since its advent in the 1990s, lead discovery has steadily evolved in response to advances in technology and processes – such as miniaturisation and laboratory automation – and the number of new screens, as well as the size and diversity of compound collections, has increased. Novartis research recognised the need for a reliable and highly automated compound management store to support standardised HTS and, between 2000 and 2002, introduced its Solution Archive (SolAr) system, combining solution storage with sample dispensing capability.

From manual to first generation automated compound management

In the 1990s, prior to the introduction of the SolAr system, Novartis maintained its liquid compound libraries in microplates stored in a cold room at 4°C and 20% humidity. Retrieval of these stock solutions was performed manually, a process that was open to human error and which had no means of tracking the volume or quality of the screening collection. With the ever-increasing number of HTS screens and the continual expansion of the compound collection, it became obvious that a move from manual to automated sample handling was necessary. Novartis adopted an automated system that combined solution storage with sample dispensing capability; the SolAr system.

The monolithic SolAr system was implemented

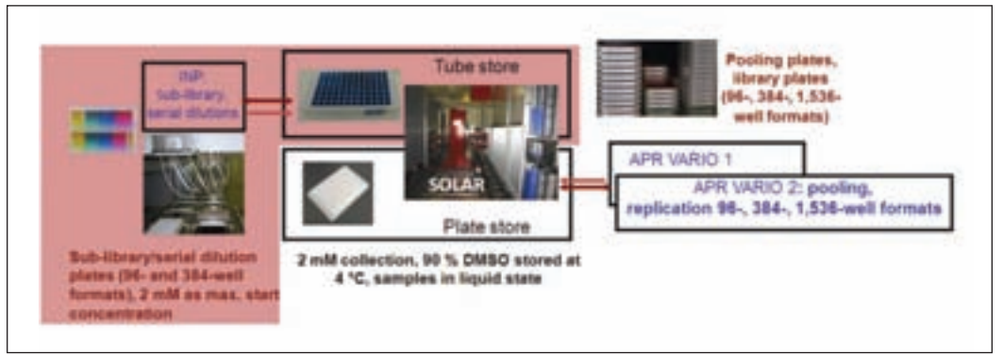


Figure 1
The NIBR SolAr system combined solution storage with sample dispensing capabilities

in 2002, fulfilling the concept of a fully automated, self-service system where an order is placed via an integrated ordering system, and the completed destination plates are collected as output. Its central liquid store, which can accommodate two million compounds, is linked to three pipetting modules – two automated plate replicators (APR) and an Individual Needle Pipettor (INP) – providing secure storage of the screening collection, as well as rapid replication of source plates and cherry-picking of individual solutions.

The SolAr storage system (**Figure 1**) is divided into two sections; a tube store and a plate store. The plate store houses a large collection of 384-well, deep well plates – the source plates for replication – connected to two CyBi®-Well vario (CyBio) APRs, enabling source plates to be replicated in 96-, 384- and 1,536-well formats for primary screening. The tube store takes advantage of REMP® Tube Technology for cherry-picking of samples, and stores the same compound library in individual tubes for a period of three years. This compound library is used for the production of validation plates. The chosen compound subset is transferred to the Individual Needle Pipettor – a Tecan Genesis RSP 150 with an eight-channel Liquid Handling (LiHa) Arm – for production of the validation plates.

Meeting the demand for increased flexibility

The SolAr system served the initially conceived HTS process very successfully but, with the evolution of integrated lead finding, its capacity and capabilities became insufficient to meet the needs of NIBR drug discovery projects. When the SolAr system was implemented, it primarily served the HTS group, delivering compound lists of up to 4,000 items requested via an integrated ordering system.

Historically, compounds were screened and validated, producing a short list for lead optimisation. Processes were standardised, requiring a limited number of different plate layouts, such as 8-dilution dose-response plates. However, the Compound Management (CM) group was increasingly handling large lists comprising thousands of compounds, as well as supporting lead optimisation activities in a lot of different laboratories, each with its own layouts and work processes.

Lead discovery at the NIBR subsequently evolved further, from standardised HTS to integrated lead finding, and the SolAr system's capabilities – just a few plate layouts and 2,000 tube movements per day – became restrictive. As working practices changed, increased flexibility in compound management was required to support lead finding strategies beyond standardised HTS, such as iterative screening, fragment-based screening and complex assay formats, for example for phenotypic or biophysical assays. Integrated lead finding is far more diverse than HTS screening, and large subsets of up to 200,000 tubes are often requested for medium throughput screening. Researchers may start with a smaller sample set, developing the assay before performing a screen, and validation lists from full deck screens are typically much larger. In addition, a range of different plate formats may be requested for complex secondary assays, requiring the implementation of far more flexible compound management processes, a flexibility that the SolAr system could not provide.

The hard-coded software used by the SolAr system also made it difficult to adapt the store to accommodate new features and requirements. While the hard link between the storage system and the liquid handling module had the benefit of enabling production overnight or at weekends, it was difficult to connect new modules, and making changes to the software was not straightforward. Programming a new destination plate layout for the INP module, for example, would also require amendments to the tube store software, which was

both time-consuming and expensive. A replacement infrastructure was needed, with the capacity and flexibility to support integrated lead finding and lead optimisation working practices.

The next generation of compound management

The demand for increased flexibility and throughput led Novartis to initiate a two-phase development of a novel, state-of-the-art automated compound management set-up to supersede the SolAr system. The first phase of the project involved replacement of the 4°C tube store and integrated INP with a two-million tube capacity -20°C store and two physically separate dose-response plate (DRP) modules, decoupling the link between the tube store and the liquid handling modules. A new approach to IT integration was also adopted. This phase is nearing completion, and the second phase of the development, the replacement of the plate store and APR modules, is currently in progress.

The second generation compound management system is quite different from the SolAr system, as all picking procedures are now performed outside the tube store. Robotic systems in the rear -20°C tube store (Brooks) transfer the selected storage trays to two separate picking stations equipped with FlexPickers™ (ABB) in an adjacent room (**Figure 2**), where the selected compounds are transferred into tube racks. Each FlexPicker performs up to 30,000 tube movements per day – the SolAr system could handle just 2,000 tube movements – at very high speed, significantly increasing sample throughput. This increase in capacity was vital to accommodate the growing compound collection and meet the demands of modern day drug discovery protocols. The picking stations are linked to movable Cytomat® C24 carousels (Thermo Scientific) by a sample transfer track with four docking stations. Once picking is complete, the tube racks are assigned a destination and transferred via the track to the specified carousel, where up to 216 tube racks can be accommodated. The tubes are then left at room temperature to thaw, assisted by fans mounted on the side and top panels of the carousel to improve the internal air flow.

Automated plate generation is performed on two customised dose-response plate modules (**Figure 3**), which are fully integrated with the Novartis LIMS. Based on the Freedom EVO® 200 platform (Tecan), each DRP module is equipped with a MultiChannel Arm™ 384 (MCA 384), a Liquid Handling Arm and a Robotic Manipulator (RoMa) Arm (all Tecan), as well as a PlateLoc® Thermal Microplate Sealer (Agilent Technologies), a Multidrop® Combi

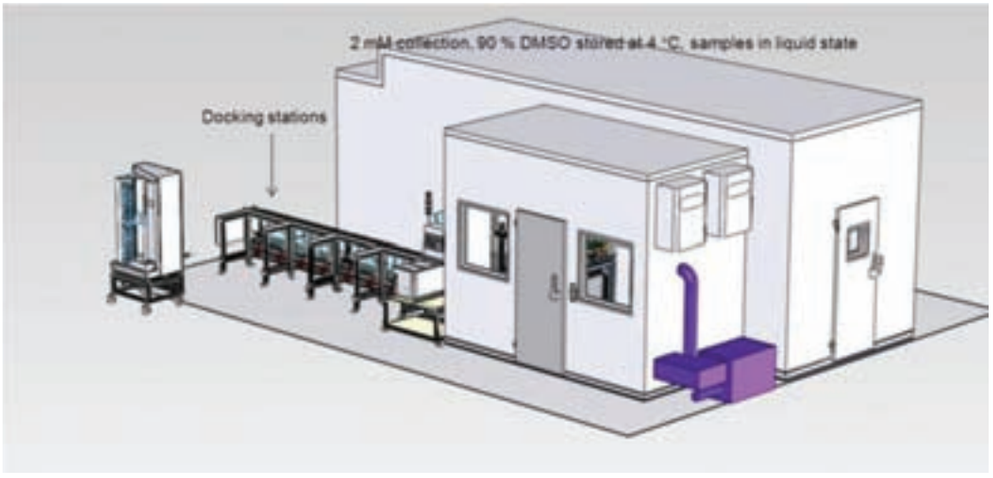
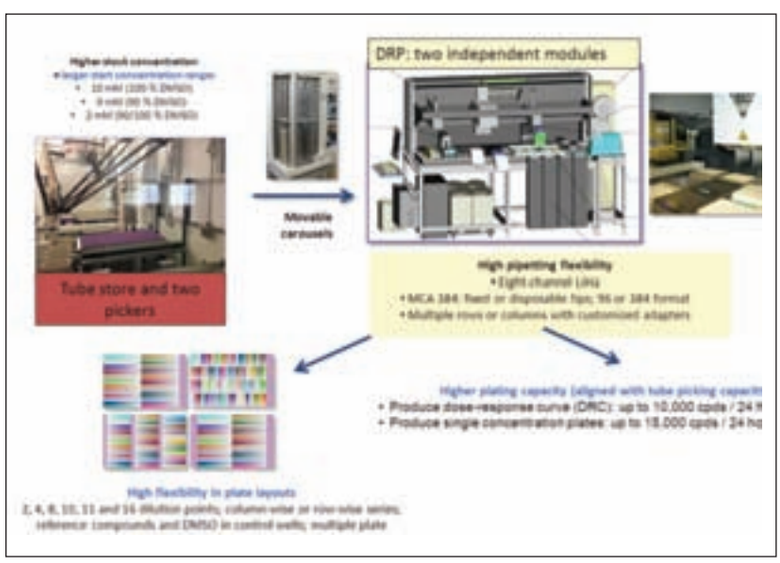


Figure 2
The new -20°C tube store houses a 10mM compound collection in 100% DMSO

nL (Thermo Scientific) for bulk filling, an argon bath, a customised 96 piercing head and a variety of MCA 384 adaptor plates (Tecan). The preloaded carousel containing the selected tubes docks to the rear of the DRP module, enabling transfer of the source racks to the work deck. Empty destination plates are stored in four stacks at the front of the DRP and moved to the deck as necessary.

Straightforward replication or stacking processes requiring single concentration plates are performed with the customised piercing head developed in collaboration with Tecan, allowing the entire compound set to be transferred in one go without the need to uncap tubes. For the production of dose-response plates, stock solutions are pipetted into rows or columns with the eight-channel LiHa, and dilution sets are prepared using various MCA adapter plates with disposable tips, selecting rows or columns as required. Backfilling of DMSO is carried out using the Multidrop Combi nL. The flexibility to use a combination of different pipetting modules, instead of relying on a single eight-channel LiHa, enables a greater range of plate layouts to be used, as well as increasing the speed of plate production. With the SolAr system, just four plate layouts were available and dose-response plate throughput was around 1,000 compounds per day. By comparison, each DRP module offers 40 plate layouts and can process 5,000 tubes per day – a huge increase in plate production.

Figure 3
The customised DRP modules offer increased flexibility and throughput



Implementation of the new tube store also provided the perfect opportunity to harmonise the lead optimisation and HTS workflows, combining the two compound collections and adopting identical stock collection and storage formats. The SolAr store housed a screening collection of some 1.5 million REMP tubes containing 2mM stock solutions in 90% DMSO, while a lead optimisation collection of about 100,000 Matrix tubes (Thermo Scientific) of 10mM solutions in 100% DMSO was maintained in a separate manual store. The new system enabled NIBR to combine these collections, standardising on a concentration of 10mM in 100% DMSO – a commonly used solvent – stored at -20°C in 2D barcoded Matrix tubes. The quality of the collection is monitored by performing systematic LC-MS measurements on all freshly prepared solutions, excluding any low quality samples from the collection. The solutions are replenished once empty, or if any signs of degradation are observed.

A flexible software solution

Upgrading the hardware is just one piece of the puzzle; any hardware is only as flexible as its supporting

software. The Compound Management group had diversified significantly since the introduction of the SolAr system, performing not only HTS screening but, increasingly, lead optimisation activities. More plate layouts were required, which meant more scripts to manage and additional plate templates to register. The NIBR IT department, in collaboration with its software development partner, GGA Software Services, based in Cambridge, Massachusetts, implemented a flexible, fully integrated, enterprise-wide software solution based on service-oriented architecture (SOA). The sample management framework (SMF) incorporates sample registration and ordering, order fulfillment, and container and plate layout registration, complemented by a global ordering system, which allows customers to check compound availability, select delivery containers, protocols and layouts, and to place orders. Once a request has been received, CM defines order priorities – lead optimisation requests must be fulfilled within 36 hours – and creates a worklist using the fulfillment software, which dispatches the information to the tube store and DRP modules for plate production. The SMF software components provide much greater flexibility, enabling CM to define and register new plate layouts and fulfillment protocols in a configuration mode, without the need for support from IT specialists.

Making the transition

The new tube store in Basel has been operational since July, and staff are now familiarising themselves with the hardware and software and optimising the workflow. Selected lead finding activities, which were previously limited by the capabilities of the SolAr system, are being supported and have already led to a doubling of the output. CM will be progressively extended to include HTS screening applications with the aim of phasing out the SolAr system next year. An equivalent tube store and DRP module set-up at the Cambridge site will become productive towards the end of 2013, harmonising processes on both sides of the Atlantic.

Conclusions

The introduction of new tube stores, DRP liquid handling modules and the flexible SMF software framework has enabled NIBR to harmonise its lead optimisation and HTS compound collections, standardising storage formats and serving integrated lead finding in a highly flexible manner. The number of available plate layouts has increased from four to as many as required and, with the capability to perform up to 60,000 tube movements and plate 10,000 compounds per day, the system has significantly improved CM's throughput. With comparable systems in use in the US and Switzerland, requests can be split more easily, while still maintaining the flexibility to accommodate specific local needs. Novartis now has global teams working on the second phase of the compound bank initiative – the replacement of the plate store and the automatic plate replicators – and, ultimately, will have plate stores and plate replication systems installed in both the US and Switzerland, completing the transition from the SolAr system to a modern, state-of-the-art platform.

Acknowledgements

The authors would like to thank the staff of the NIBR IT and Compound Management/Engineering groups, whose involvement in the compound bank initiative helped to ensure its successful implementation, and to acknowledge the technical advice and support of Urs Zuehlke, Georg Filipovic and Ralf Masantschek from Tecan. DDW

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