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Innovative Microarray Solutions

Largest Human Protein Array (CDI's HuProt™) Printed with Arrayjet Inkjet Bioprinting Technology

Arrayjet Advance™ manufacturing services have enhanced protein array production and improved yield while minimising sample wastage for a US-based proteomics company

Issue

The customer was experiencing frequent production delays, loss of yield, batch inconsistency and sample wastage with their previous pin-spotting technology. They were struggling to effectively commercialise their product and maintenance costs were exponentially increasing. The low throughput contact pin-printing system was unable to efficiently deliver high quality protein arrays. Consequently, the US-based proteomics company approached Arrayjet for their in-house expertise in inkjet protein printing.

Arrayjet Advance™ Services

The CDI project was supported through Arrayjet Advance™ custom printing services, which Arrayjet launched in 2011. This end-to-end CRO microarray service allowed the facility to access application support through the wealth of scientific expertise and microarray knowledge at the Arrayjet HQ. The aim was to utilise Arrayjet's custom printing services to successfully transfer, optimise and commercialise their protein assay.

Technology transfer

The Arrayjet Advance™ services were also used to deliver efficient manufacturing of the human protein library consisting of >19,000 GST fusion proteins probed with anti-GST antibody in duplicates for high throughput array production. The project was carried out on epoxy and nitrocellulose surfaces at 4 °C with Arrayjet's Ultra-Marathon II microarrayer and Jetmax™ environmental control unit. Multiple print runs were performed to determine the most optimum printing conditions and highest signal intensities.

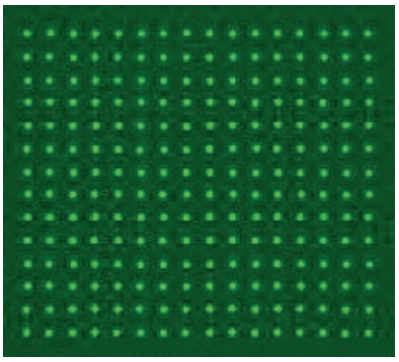

Figure 1: Assay optimisation onto epoxysilane slides

Assay Optimisation

A small subset of protein samples was printed onto epoxy and nitrocellulose surfaces using Arrayjet's Ultra-Marathon II microarrayer at 4°C. Various test volumes were dispensed at a 165µm spot pitch to determine optimum spot deposition volumes and spot diameter. 14 identical miniarrays were successfully printed with over 99% spot circularity at 100pL spot volume (figure 1). Highly accurate data was obtained from minimum sample volume.

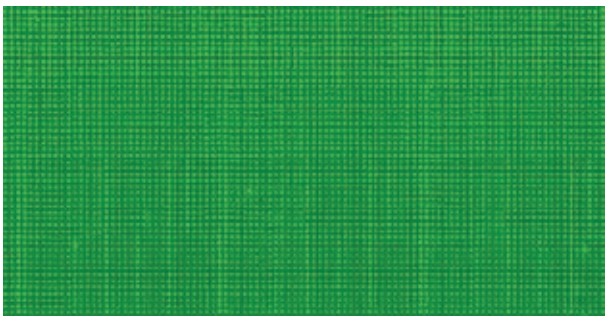

Figure 2: Small subset of human protein samples printed across 200 identical Grace BioLabs nitrocellulose slides

Inkjet batch printing

A larger subset of protein samples was then printed onto 200 epoxy and nitrocellulose glass slides to analyse spot reproducibility, circularity and morphology as shown in figure 2. Sample wastage and evaporation was minimised using Arrayjet's specially designed Jetguards™ overlaid on the source plates.

"CDI Laboratories purchased an Arrayjet 'Ultra Marathon II' to print microarrays for our 'HuProt' product line, focused arrays with smaller subset of proteins for discovery arrays and hybridoma project work. CDI are planning to print monoclonal arrays and membrane protein arrays with the new technology in the near future. Arrayjet's technology was chosen as we needed to quintuple capacity, preferably with one Instrument, its hi-tech, precise and user friendly capabilities was an obvious benefit. One of the added advantages of the company structure is that they provide in-house printing through 'Arrayjet Advance' and we were able to carry out several studies over several months to reaffirm our belief that the technology would improve many facets of our operation."

Dr. Ignacio Pino, CEO, CDI Laboratories

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High Density printing

The protein assay was successfully transferred from the previous pin-spotting platform over to Arrayjet's high speed inkjet printing system utilising the expertise of Arrayjet Advance™ services. The next step was to evaluate high-density printing capabilities of the Ultra-Marathon II microarrayer. A section of human protein library was printed onto epoxy slides to evaluate batch consistency and feature variability. High density printing was carried out to deliver reproducible hexagonal arrays onto a variety of substrates whilst lowering background and CV values. The results showed no spot merging and consistent spot alignment.

Human Protein Library Printing

An entire library of human proteins consisting of over >19,000 GST fusion proteins probed with anti-GST antibody was printed in duplicates at 200pL spot volume, across a single batch of 500 identical slides (figure 3). As per the agreed success criteria, >97% of the spots were present on all the slides with a CV value <20% representing satisfactory intra-slide batch consistency and A spot circularity of over 90% across all arrays. Validation of results and functional analysis of the arrays was performed by the customer at their facility and results were positive and highly accurate.

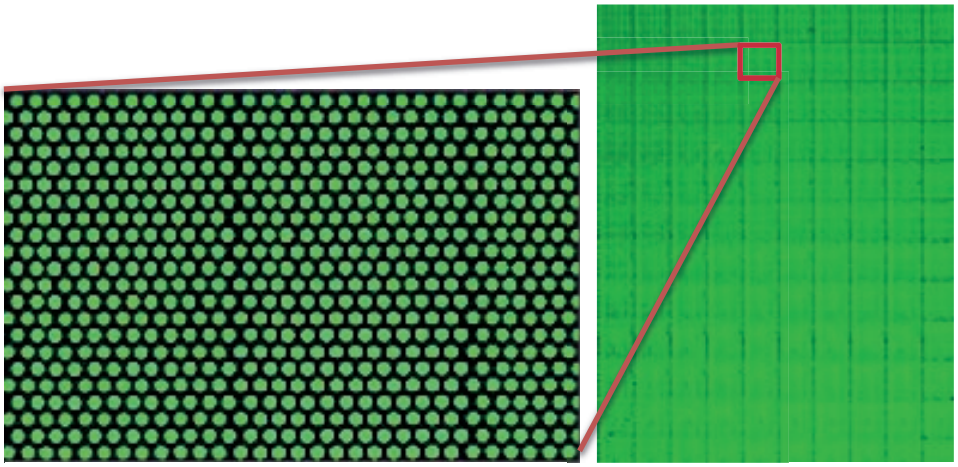

Figure 3: Largest Human protein library printed onto Grace Bio-Labs PATH protein microarray slides

Conclusion

The US-based proteomics company located in Baltimore, CDI Laboratories, is now able to successfully produce the largest human protein microarray in the world. Their HuProt™ Version 3.0 arrays are being spotted across 1000 slides in each batch. The company has purchased an Arrayjet Ultra-Marathon II to perform efficient in-house analyses of thousands of protein interactions using minimal amounts of valuable clinical samples. The Arrayjet Advance™ service and non-contact piezoelectric technology has significantly increased their production efficiency for printing large quantities of protein microarrays. The environmental control unit also enables retention of the native protein conformation with utmost precision and maintains result consistency whilst sustaining high quality spot morphology and array alignment.

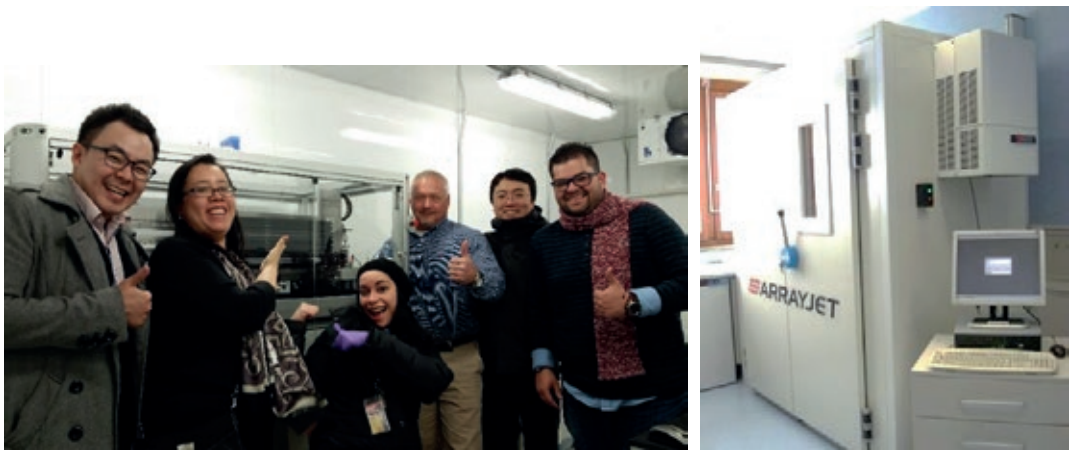

**Figure 3: Left - Team of extremely satisfied Arrayjet customers after Ultra Marathon II installation in Baltimore, USA.
Right – Arrayjet Ultra Marathon II system enclosed within the JetMax™ environmental control unit for low temperature printing.**

"Arrayjet's Jetspyder™ sample aspiration device, effectively eliminates any cross contamination between protein samples, so the quality of our slides is significantly improved. Previously we could produce just over 150 slides of acceptable quality in a print run, too slow a pace for the scale and growth of current and future demands; being able to print up to 1000 slides quickly, in a highly efficient manner was incredibly appealing to us."

Dr. Heng Zhu, Professor Johns Hopkins School of Medicine