Technical Note

UHN Microarray Centre

Corning UltraGAPS™ slides to replace CMT-GAPSII™ slides for all UHNMAC microarray products

- Following successful validation, all production batches will be printed using UltraGAPS™ slides (Corning Inc., New York)
- These slides provide better signal to noise ratios, have less background, and require less post-printing processing than the slides used previously
- Prior to using UltraGAPS™ slides, the UHNMAC used Corning CMT-GAPS™ and CMT-GAPSII™ slides for the manufacture of cDNA and oligonucleotide microarrays

Introduction

Since 1998, the University Health Network Microarray Centre (UHNMAC) has been printing and distributing ready-to-use microarrays to over 700 research labs worldwide. Over the years, we have evaluated different coating chemistries (amine, epoxide, aldehyde, etc.) and commercial suppliers have developed new coating surfaces to improve performance and consistency. The UHNMAC has always printed arrays using commercially available microarray slides for consistent, reliable performance. Another benefit of using commercial slides is that many are barcoded affording the ability to track each printed array.

In 2003, UltraGAPS™ slides (Corning Inc., New York), which promised more consistent quality, were successfully validated in our lab. At that time, UHNMAC array products were printed on Corning CMT-GAPS II™ slides and prior to these slides were printed on Corning CMT-GAPS™ slides. The proprietary gamma-amino silane coating of UltraGAPS™ slides provide a more hydrophobic surface than CMT-GAPS II™ slides which allow for higher density arraying and more consistent spot morphology.

Besides improved performance, the other advantage with the UltraGAPS™ slides is there is minimal post-printing processing involved (simply UV cross-linking). Processing CMT-GAPS II™ slides involved the use of hazardous chemicals such as succinic anhydride and n-methyl-pyrrilidinone (M2P). These additional processing steps sometimes introduced slide variation from lot to lot, especially when stored inadequately during shipping and handling. One of the more common problems encountered was high fluorescent background even with optimised washes after hybridisation. The only disadvantage of switching to the UltraGAPS™ slides for manufacturing was their increased cost.

Method and Results

The UltraGAPS™ slides were evaluated and the results from preliminary experiments showed improvements over the CMT-GAPS II™ slides. Further studies were done to optimize the post-printing processing protocol. Unlike CMT-GAPS II™ slides, which were treated with succinic anhydride and M2P to prevent high background fluorescence, UltraGAPS™ are simply UV cross-linked after printing to ensure the spotted nucleic acid would remain on the slide during hybridisation.

A comparison of the Mouse15k clone set printed on a) CMT-GAPS II™ (UV cross-linked and processed with M2P) and b) UltraGAPS™ (UV cross-linked only) is shown (Figure 2). This image indicates that UltraGAPS slides with UV cross-linking alone, provides

a better image in terms of consistent spot morphology and low background fluorescence.

Discussion and Conclusion

Upon evaluating UltraGAPS™ slides, it was discovered that treatment with hazardous M2P was not necessary, nor was it necessary to wash the slides in a SDS solution. By eliminating these post-printing steps, the time and cost savings offset the additional cost of the UltraGAPS™ slides. Another benefit of eliminating the chemical processing is that the salt from the spotting solution remains on the slide and allows for the visual inspection of each microarray (Figure 1).

The UHNMAC currently uses UltraGAPS™ slides to print all cDNA and oligonucleotide microarrays manufactured in-house. It is recommended that UHNMAC printed arrays, which are shipped ready-to-use, are stored in dry place and protected from light. Prior to packaging and shipping, printed arrays are stored in a desiccator in the dark. The recommended shelf life for our printed arrays is between 6-8 weeks under proper storage conditions.

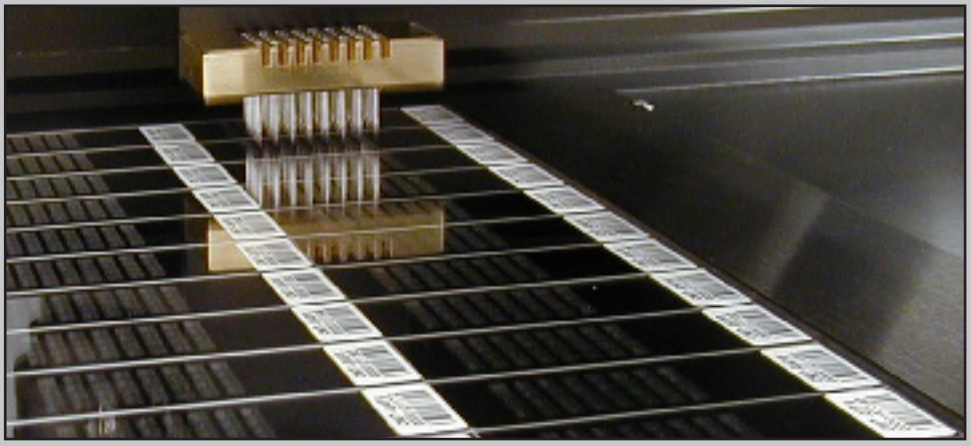

Figure 1. During array manufacture, the 3X SSC spotting solution dries on the slide and allows for the visual inspection of each microarray post-printing. The benefit of eliminating the chemical processing is that the salt remains on the slide. During chemical processing, the salt is removed and the DNA spots become invisible.

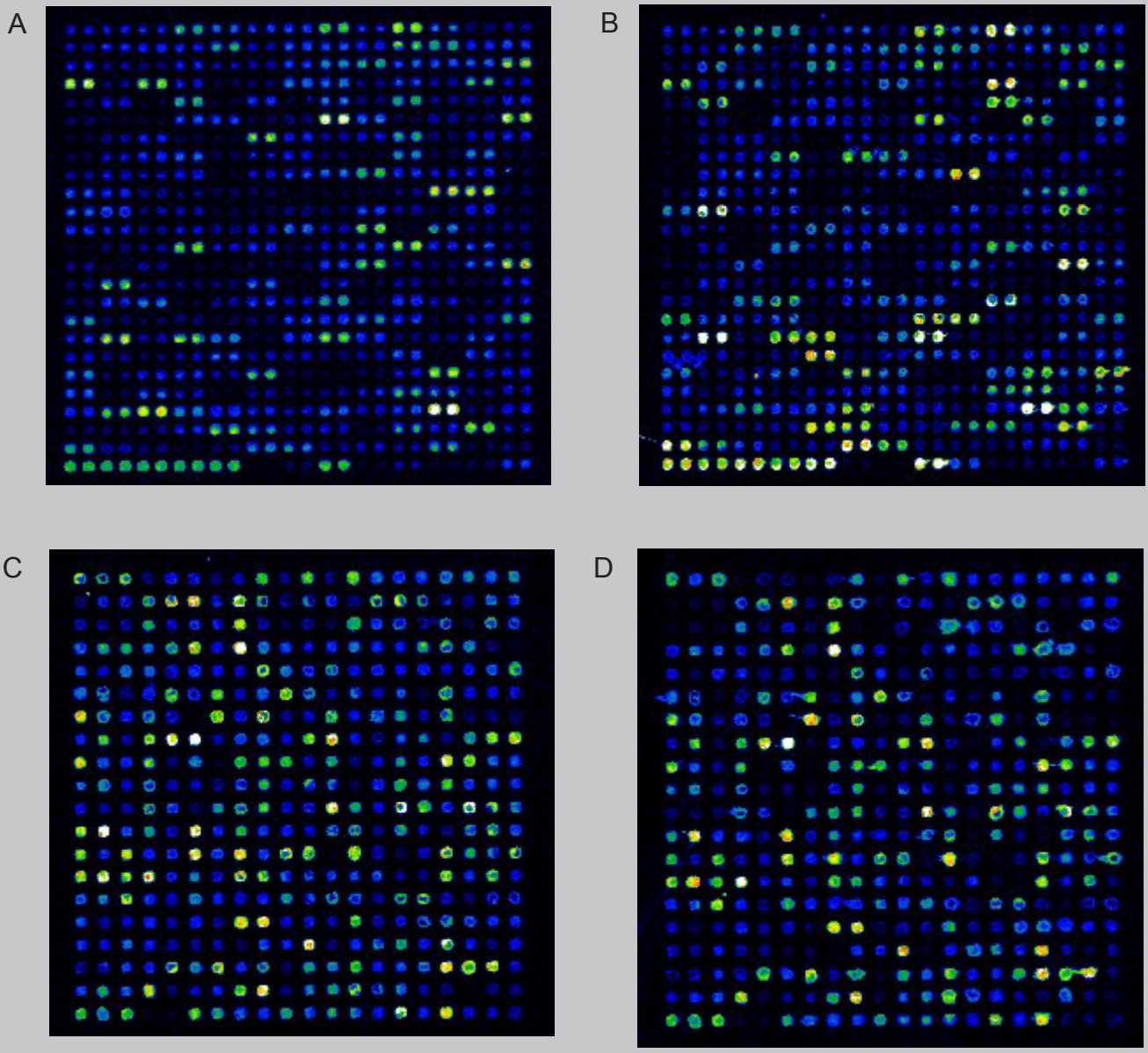

Figure 2. Top Images: A comparison of the Mouse15K cDNA clone set printed on an UltraGAPS™ slide (UV cross-linked only; image A) and CMT-GAPS II™ slide (UV cross-linked and processed with M2P; image B). Bottom Images: A comparison of the Human19K clone set printed on an UltraGAPS™ slide (UV cross-linked only; image C) and an UltraGAPS™ slide (UV cross-linked and processed with SDS washes after printing; image D). UltraGAPS™ slides that are only UV cross-linked after printing (image A and image C) provide the best quality image in terms of consistent spot morphology and low background fluorescence.
All images are of cyanine5-labelled Universal Human Reference RNA labelled following the UHNMAC standard direct-labelling protocol.