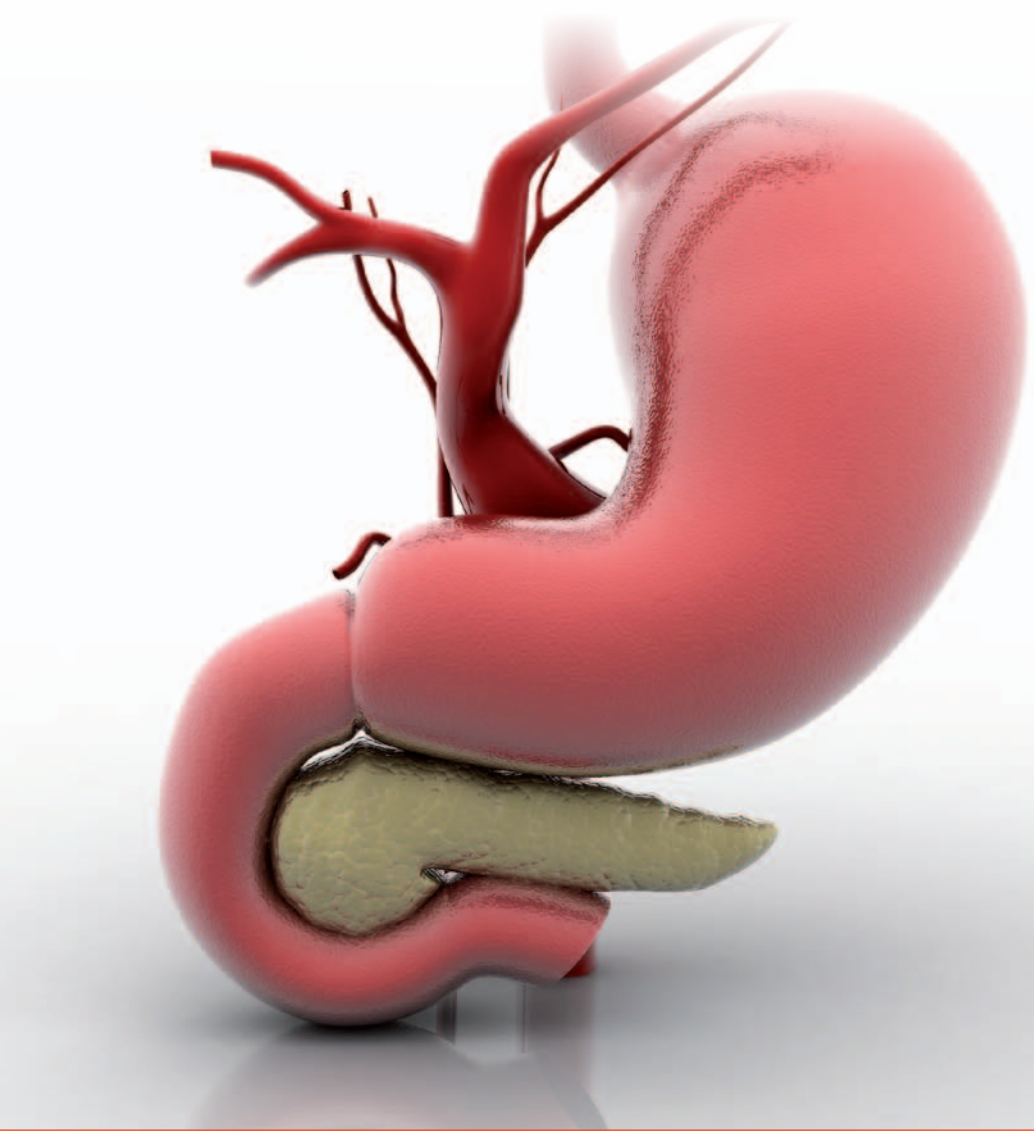
Developing a proteomic fingerprint for pancreatic cancer

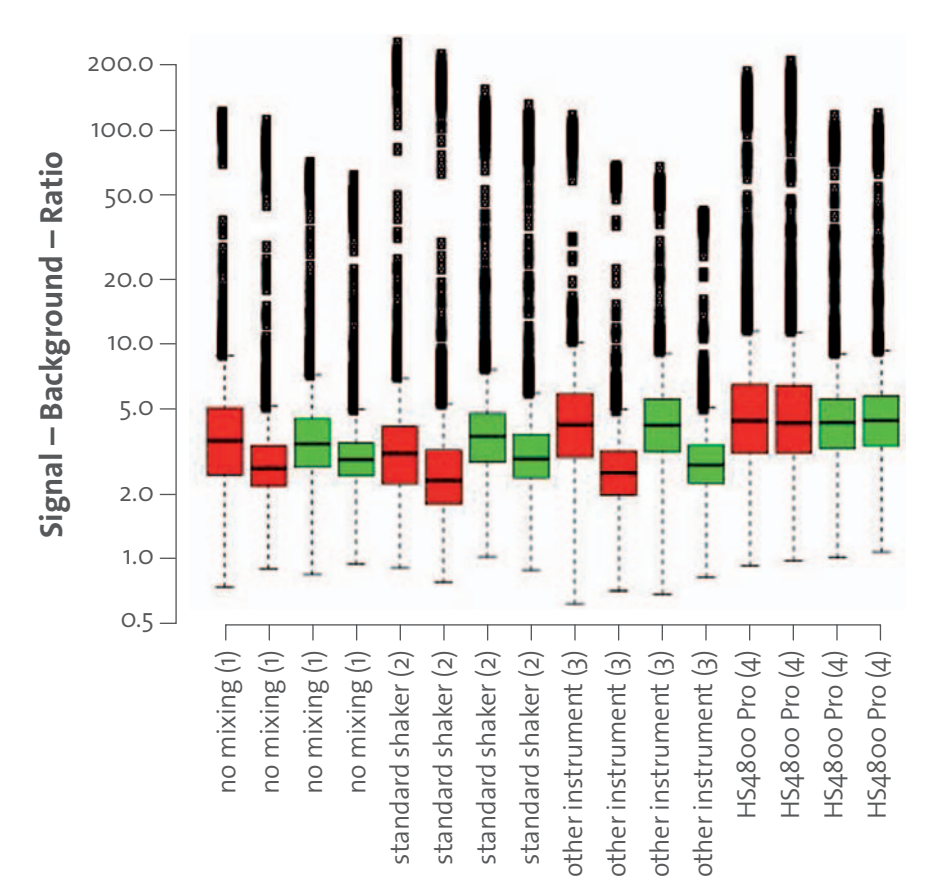
Scientists at the German Cancer Research Center are using an HS 4800™ Pro microarray hybridization station and a PowerScanner™ to investigate pancreatic cancers. By combining these automated solutions with in-house developed antibody microarrays, the Center is able to study pancreatic cancer at a protein level, helping to better understand the basic biology of the disease and develop new screening strategies for early detection.



Pancreatic cancer has a very high mortality rate as symptoms can go undetected for many years before diagnosis, limiting the possibility for treatment. To help alleviate this problem and improve survival rates, the Deutsches Krebsforschungszentrum (DKFZ, the German Cancer Research Center) is using protein microarray techniques to help develop early diagnostic and prognostic markers. Dr Jörg Hoheisel from the DKFZ's Division of Functional Genome Analysis, explained: "Our group is focused on the development of new techniques which can be applied to routine diagnostic applications or can help to understand the complex biology of pancreatic cancer. We have had a strong interest in microarray technologies for a long time and moved into the proteomics field about a decade ago, developing a number of peptide, protein and antibody microarrays to look at various aspects of the disease."

Dr Christoph Schröder continued: "One of our major projects has been the development of an antibody microarray targeting over 740 proteins of interest in pancreatic cancer[1]. The target proteins are generally either strongly up- or down-regulated in tumor cells, and this microarray is designed to allow the investigation of the key cellular pathways involved in cancer, as well as to help identify biomarkers of diagnostic or prognostic value. Throughput and reproducibility are crucial to enable the generation of meaningful data in this type of application. We have been using an automated microarraying platform and a system with automatic mixing during incubation for some time, and have been able to achieve very good results. However, all washing and drying steps were performed manually, and so we looked at the possibility of further automation of our workflow to increase throughput and further improve reproducibility."

"We trialed hybridization stations from various manufacturers, and the Tecan HS 4800 Pro provided the best solution for our needs," Christoph said. "Automation of the washing and drying steps offered the



Signal-to-noise (S/N) ratios for multiple replicates of antibody microarrays representing 741 different cancer-related proteins. Incubations were performed in quadriPERM® chambers:

(1) without mixing

(2) with mixing on a standard lab shaker

(3) with an alternative commercial system for microarray incubation, in all three cases using manual washing and drying steps, or

(4) fully automated using the HS 4800 Pro.

Red bars depict the S/N ratios for the red fluorescence channel, and green bars correspond to the S/N ratios for the green channel

robustness and low variability we needed, as well as improving throughput. It also significantly improved the signal-to-noise ratio for our arrays, allowing us to reduce the incubation time from 16 hours to just 1-3 hours without compromising the quality of results, which increased our throughput even more."

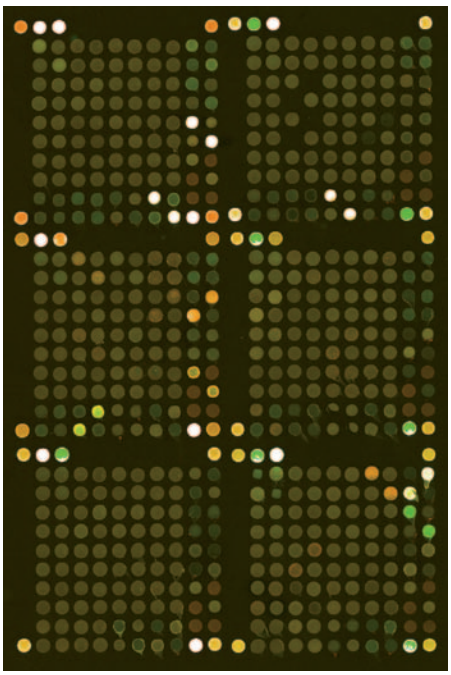
"As we mainly work with clinical samples – both from hospitals here in Germany and as part of larger European projects – the sample volume required for analysis was another important consideration. Only very limited amounts of biological material are available, and samples cannot usually be replaced if an experiment fails. The HS 4800 Pro combines excellent reliability with very good reproducibility; we get the same data from any given sample, even if it is run on different days. As our throughput was increasing rapidly, we also looked at replacing our old microarray scanner. Again, we looked at instruments from several manufacturers, and felt that Tecan's PowerScanner was the best fit for our laboratory. The system is intuitive to use, and its robust design gives you confidence in the results."

Christoph concluded: "This combination of instruments offers excellent reproducibility, and we are in the process of publishing several studies based on the data we have generated. We have also recently begun a collaboration based on this technology with a group at McGill University in Montreal, Canada, using the same instruments, microarrays and protocols in both laboratories to extend the use of the antibody array platform. Overall, we have been very pleased with the performance of the Tecan instruments. The biomedical results we have achieved have led to the creation of a spin-off company focusing on the development of diagnostic tests and custom services based on antibody microarray screenings."

1. Schröder, C *et al*. Dual-color proteomic profiling of complex samples with a microarray of 810 cancer-related antibodies. *Mol Cell Proteomics*, 2010, 9, 1271-80.

To find out more about Tecan's HS 4800 Pro and PowerScanner, visit **www.tecan.com/microarray**

For more information on the DKFZ's Division of Functional Genome Analysis, visit **www.dkfz.de/funct_genome**



Dual-color image of an antibody microarray incubated with two different plasma samples