Institution: University of Abertay Dundee

Unit of Assessment: 05

Title of case study: Development of a novel light-scattering instrument and applications for measuring molecular interactions and aggregation analysis

1. Summary of the impact (indicative maximum 100 words)

Professors Zhelev (UoA5) and Bradley (UoA15) explored the scope and demonstrated the feasibility of using light-scattering methods for quantitative analysis of macromolecular associations and aggregation, including protein-protein and protein-DNA interactions. 16 years of design and development research was translated into a marketed product – the PAM™Zero - a novel hand-held, low-cost protein aggregation monitor capable of detecting macromolecule aggregation in microliter sample volumes. Manufactured and sold through a spinout company, Norton Scientific Inc. (established in 2010 and valued at $7M), this portable instrument is used in commercial Quality Control and academic research and has been sold to a range of stakeholders e.g. drug development companies, for food safety and water pollution monitoring.

2. Underpinning research (indicative maximum 500 words)

Zhelev pioneered research, which indicated novel application of laser light scattering for studying protein-protein and DNA-protein interactions [1]. The concept was informed by technology normally used by chemists to, for example, quality control the manufacture of nanoparticles. In a study conducted in 1996 in collaboration with Protein Solutions Inc., Zhelev, then a consultant research scientist at the Randall Institute, King's College London, demonstrated the efficacy of dynamic light scattering to measure protein-protein and DNA-protein interactions in vitro. In one of the first publications proving efficacy of light scattering methods for protein aggregation assay, he used as a model system the bHLH zip protein Max, purified as a recombinant GST-fusion and chemically synthesized target DNA containing Max binding sequence. This underpinning research [1] concluded that dynamic light scattering offered assay speed and sensitivity, making it an attractive alternative to the then available biophysical and biochemical protein-protein assay methods used in biotechnology (such as surface plasmon resonance, analytical ultracentrifugation, two hybrid system, phage display and chromatography) which are expensive, time consuming, and demand highly skilled technicians.

In 1997 Zhelev moved to Dundee and continued work on laser light scattering applications in biotechnology in collaboration with Bradley (an expert in the design, development and implementation of mechatronic devices and systems at the University of Abertay Dundee) and White (Founder and Chief Scientist of Optokem Ltd. and later RC2 Inc., a company relocated from Bangor to Dundee developing and supplying bench-top light scattering instrumentation), in particular, they developed the underlying technologies and validated the concept in relation to applications in biochemistry. In 2003 Zhelev was appointed as Director of the Centre for Molecular and Cellular Biosensor Research (CMCBR) at the University of Abertay Dundee, where he explored light scattering methods for quantitative analysis of macromolecular associations as part of a wider cancer biology research programme. Zhelev's research expertise and insight clarified to him the limitations of existing light scattering technologies which required large sample sizes >1ml. He and other modern-day molecular biology researchers required rapid, reliable, low-cost, easy to use screening tools for measuring protein interactions within microlitre sample volumes. In 2005 Zhelev published a theoretical review discussing the underpinning theoretical science and prospective applications of light scattering for the characterisation of biomolecules [2].

Zhelev therefore initiated an innovation programme with Bradley and White to translate these ideas into a novel commercially viable measurement system for biotechnology research. The resulting PAM™Zero, a small, hand-held, advanced protein aggregation instrument allowing sensitive (particularly for 10-100 kilodalton proteins), easy, rapid, low-cost screening of small (1-2 microlitre) recoverable aliquots (3, 4), was launched in 2011 by Norton Scientific Inc. Zhelev's key contribution was to define, develop and validate the workflow methodology (including identifying constraints to sample preparation), and identify key applications for protein research including crystallization and the study of aggregation kinetics [4]. This 8 year on-going collaboration has now completed the design specifications for a follow up – the PAM 2.0, incorporating additional functionality (see section 4 for details).

3. References to the research (indicative maximum of six references)

1. Zhelev N., Buckle R., Snyder D. and Marsh P. Studying of protein-DNA binding by dynamic light scattering. ***Cell. Mol. Biol. L.*** (1996) vol. 1(2), pp 199 – 203.
2. Zhelev N. and Barudov S. Laser light scattering applications in biotechnology. ***Biotechnol. & Biotechnol. Eq.*** (2005) vol. 19 (3) pp 3 – 8.
3. *White R, Zhelev N, and Bradley D. Issues of Innovation, Design and Development in Relation to the Implementation of Light Scattering Instrumentation. In ***Mechatronics and manufacturing engineering*** (J.P. Davim ed.) Woodhead Publishing, Cambridge, (2011) pp 1 – 45.
4. Zhelev N, Khalil HS, and White R. New approaches to rapid screening for protein aggregation. ***Biotechnol. & Biotechnol. Eq.*** (2013) vol. 27(6) (in press)

* Review included as it explores the innovation methodology used for the engineering technology involved.

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4. Details of the impact (indicative maximum 750 words)

Research by Zhelev on the application of laser light scattering technologies for protein and DNA analysis, supported by the work of Bradley on the mechatronic principles underlying the design and manufacture of specialist measurement systems (both at the University of Abertay Dundee), has resulted in the development and commercialisation of a new instrument for use in the pharmaceutical and biotechnology arena. This novel hand-held instrument, the PAM™Zero, was launched in 2011 by Norton Scientific Inc. generating sales exceeding $100,000 in the first 6 months (http://www.norsci.ca). Further interactions with Norton Scientific Inc. following this success has led to an on-going project defining the workflow for follow-up future products and has already completed design specifications for a follow up – the PAM 2.0, incorporating additional functionality.

Zhelev's research and deep understanding of the measurement requirements for a suitable light scattering system appropriate for use by molecular biotechnologists, was critical in informing the design and development of an essentially new approach to laboratory measurement of protein interaction. He focussed on improving the speed and cost efficiency of drug development research

rather than on the creation of new theoretical techniques of measurement. Previously available methods allowing protein-protein and protein-DNA interactions assay were costly, time-consuming and demanded highly skilled operators. Alternative, laser light scattering technologies available in the 1990's were expensive bench-top devices requiring large sample sizes (>1ml) rendering most unsuitable for molecular biotechnology studies. Zhelev and Bradley's informed approach to the design and development of a technology suitable for studying protein interactions was to be guided by the requirements of research and development scientists (speed, sample size, measurands, ergonomics).

Research at Abertay was undertaken in the following key areas:

- Identification of the most appropriate applications for use of rapid screening by light scattering of protein and DNA interactions, including crystallization and aggregation kinetics;
- Biochemical analysis to define, develop and validate the workflow methodology (including constraints on sample preparation) which eventually formed the basis of the Operating Manual;
- Ergonomic design of the detector sub-system taking into account factors of crucial importance to a protein biochemist, such as minimising sample loss (samples are recoverable), maximising sensitivity and ease of data interpretation;
- Design issues associated both with the operation of the instrument and its manufacture. This concentrated on two main areas (i) the ability to automate various aspects of the process associated with the measurement process in order to increase throughput and (ii) the use of manufacturing technologies such as 3D printing to customise specific system elements.

As a result of this research work, which also involved testing at Queens University in Kingston, Ontario, a prototype detector using fibre optics and an ultra-low volume flow cell was developed and tested for use in protein crystallography and protein aggregation kinetics.

This research led directly to the formation of a new spin out company, Norton Scientific Inc, founded in Ontario Canada in 2010 (to improve access and penetration of the US marketplace) and supported by a Science Advisory Board that included Zhelev and Bradley as founder members (with White). Further interactions with the company after testing of the prototype resulted in a major improvement in sample handling by creating a novel sample head capable of measuring protein samples within a pipette tip, thereby eliminating sample waste. Norton Scientific, following launch in 2011 of the resulting detector called PAM™Zero, obtained a private listing on the Frankfurt Borse First Quotation Board, valuing the company at $7M by late 2012. Early sales (globally) of the PAM™Zero exceeded $100,000 in the first 6 months from launch, and has helped diminish the effect of analytical bottlenecks delaying the launch of new drugs by biotechnology and pharmaceutical R&D.

Since launching the first version of the PAM™Zero, it has penetrated a number of key research and development markets in North America, India/Asia and Europe, selling to a mix of drug development companies, the environmental and food and drink industry and to academic research groups studying protein structure. Early feedback from key customers has already been consolidated into the design of a new PAM (the PAM 2.0) that will incorporate a UV/Vis spectrophotometer option, (developed with the support of Bradley) and incorporating novel 3-D printing technology in its manufacture, to be developed in collaboration with the University of

Abertay Dundee. This functionality will produce a slight increase in cost but increase the usability of the instrument significantly.
The applications research undertaken by Zhelev has provided the foundation for a number of collaborative research links established by Norton Scientific since 2010, including Queens University, Kingston, Canada and Harvard Medical School, USA. These links have also extended the research network of the University of Abertay Dundee. As a result of this work, new tools that can accelerate drug discovery research, have and will continue to be developed and commercialized for the bio-pharmaceutical and –technology markets.

5. Sources to corroborate the impact (indicative maximum of 10 references)

1. Norton Scientific Inc. website: http://www.norsci.ca
2. Norton Scientific Inc., Founder & Chief Technical Officer can discuss the involvement of Zhelev & Bradley in the innovation process and may provide some confidential information related to sales. User details may be provided by Norton Scientific Inc. if required once applicable data privacy regulations have been adhered to and under the auspices of a mutual NDA with NSI.
3. Norton Product Flyer and customer presentation using examples of data produced by Zhelev. This confirms and illustrates the involvement of Zhelev in the development of PAM™Zero : http://www.norsci.ca/norton-scientific-to-collaborate-with-prof-zhelev-head-of-cancer-systems-biology-at-university-of-abertay-dundee-uk-on-the-development-of-protein-protein-interaction-applications
4. Company value - $7M by late 2012, information can be obtained from Norton Scientific Inc. Founder & Chief Technical Officer.