Institution: Aberystwyth and Bangor Universities - Biosciences, Environment and Agriculture Alliance (BEAA)

Unit of Assessment: 6: Agriculture, Veterinary and Food Science

Title of case study: Improvements in biomass measurement have changed practitioner approaches and improved profitability in commercial brewing and biotechnology

1. Summary of the impact

BEAA research has impacted positively on commerce in brewing and biotechnology companies worldwide through continuous collaboration with Aber Instruments, an AU spin-out company formed to commercialise university research. Aber Instruments has supplied over 1000 fermentation monitoring systems world-wide for the on-line measurement of viable biomass concentration, providing improvements in speed and accuracy over previous off-line, culture or stain-based procedures. On-line, real-time monitoring of viability during fermentation reduces costs and improves product quality, leading to practitioners in large breweries including Anheuser Busch, SABMiller, Inbev, Coors, Diageo, Heineken, Suntory and San Miguel adopting the Yeast Monitor as part of their standard operating procedures. The new Futura instrument, which utilises the same technology developed from BEAA research, was launched in 2009 and is now used by major biotechnology companies including Genetech, Novo, Biogen Idec, GlaxoSmithKline, Centocor, Sandoz, Eli Lilly and Genzyme to monitor biomass in a much wider range of fermentations.

2. Underpinning research

In brewing, the quality of the raw materials impacts directly and substantially on the quality of the finished product, and thus on the profits and reputation of the manufacturer. The most difficult aspect to control is the yeast – as a biological agent both the viability and vitality of the organism must be monitored to maximise efficiency and product quality. As yeast is recycled from one brew to the next following compaction, cold storage and acid washing, its quality can vary considerably. Standard microbiological methods such as plate counts or growth in liquid culture provide only *a posteriori* information about the yeast quality – by which time the brew may have been spoiled (leading to loss of profit, environmental issues if it requires disposal or storage costs if it is to be blended). Historically, stain-based methods have been used to monitor viability but these introduce errors through sampling from a heterogeneous suspension with associated contamination risk and increase labour costs and delays associated with processing of the off-line sample and data.

The basis of the biomass monitoring systems developed through BEAA research and commercialised by Aber Instruments is the real-time and on-line radio-frequency impedance measurement of viable cell concentration. Following on from earlier AU work, since 1993 research has included analysis of brewer's yeast in laboratory conditions and also under industrially-relevant challenging situations such as at high volume fractions [3.1a] with the presence of non-biological particulates, fluctuating or biologically-extreme temperatures, presence of gas bubbles [3.1b], complex and variable growth media, etc. Thorough analysis of the measuring electrodes and electronics [3.1c,d,e] has led to major improvements in the quality and reliability of data obtained using this methodology and these advances are the direct result of AU patents assigned to Aber Instruments [3.2]. Additionally, the sensitivity of the method to loss of viability, changes in cell size during growth or nutrient limitation, aggregation of cells, fouling of electrodes etc have all been investigated within BEAA [3.1a-e, 3.3a,b]. The publication of this important and underpinning research has assisted the company in improving and verifying the robustness of the technique and has shown it to be a method that is suitable for real-world applications. This has contributed to the acceptance of this technique as a standard approach by practitioners within the brewing industry.

In addition to work with brewery-relevant samples, BEAA research has included a wide range of other biological material including bacteria, yeast, mammalian and plant cells, all at industrially-relevant volume fractions [3.1a]. This has led to wider applicability of the technique within the

biotechnology sector (for example the technology has been adopted by Genetech, Novo, Biogen Idec, GlaxoSmithKline, Centocor, Sandoz, Eli Lilly and Genzyme). BEAA have also provided expert advice on potential competitor patent applications protecting the interests of Aber Instruments.

Currently, EU-funded research is further developing the accuracy and the amount of additional biologically relevant information that can be obtained from online monitoring, while DEFRA, BBSRC and ESDF (Welsh Government) -funded projects are seeking to extend the methodology to biofuel applications through monitoring degradation of plant material.

The original work was initiated within the research group led by Professor Douglas Kell at Aberystwyth University. He remained active in this area as a member of professorial staff until his move to Manchester University in 2002. Dr Hazel Davey has taken an increasing lead in this research as a member of staff since 1994, initially as a PDRA, Research Fellow, Lecturer and now as a Senior Lecturer. She currently supervises three postgraduate students who contribute to this research area. Dr Christopher Davey is co-author with Douglas Kell of the most recent patent [3.2] and wrote two books that are still supplied by Aber Instruments as the standard reference for this technology [3.3 a,b]. He is now a researcher on an open-ended contract at AU and continues to contribute in particular to data analysis and interpretation of biomass signals.

3. References to the research

3.1 Research papers: Over 20 including:

a) Davey, C.L., Davey, H.M., Kell, D.B. and Todd, R.W. (1993) Introduction to the Dielectric Estimation of Cellular Biomass in Real Time, with Special Emphasis on Measurements at High Volume Fractions. Analytica Chimica Acta, 279, 155-161. *Extends the applicability of models that relate capacitance to biomass concentration by explaining and adjusting for loss of linearity of signal at very high biomass concentrations. This is key to enabling the methodology to be applied to high volume fraction yeast slurries used in pitching at the start of fermentations and to solid substrate fermentations.* http://dx.doi.org/10.1016/0003-2670(93)85078-X

b) Davey, H.M., Davey, C.L., Woodward, A.M., Edmonds, A.N., Lee, A.W. and Kell, D.B. (1996). Oscillatory, Stochastic and Chaotic Growth Rate Fluctuations in Permittistatically-Controlled Yeast Cultures. Biosystems, 39, 43-61. *Demonstrates the use of radio-frequency impedance for on-line monitoring biomass concentrations in real-time, allowing automated adjustment of nutrient flow rates to control the biomass concentration*. http://dx.doi.org/10.1016/0303-2647(95)01577-9

c) Davey, C.L. and Kell, D.B. (1998). The Influence of Electrode Polarisation on Dielectric Spectra, with Special Reference to Capacitive Biomass Measurements. I. Quantifying the Effects on Electrode Polarisation of Factors Likely to Occur During Fermentations. Bioelectrochem. Bioenerg. 46, 91-103. *Explain the theory and practical application of the research behind the above patent*. http://dx.doi.org/10.1016/S0302-4598(98)00131-7

d) Davey, C.L. and Kell, D.B. (1998). The Influence of Electrode Polarisation on Dielectric Spectra, with Special Reference to Capacitive Biomass Measurements. II. Reduction in the Contribution of Electrode Polarisation to Dielectric Spectra Using a Two-Frequency Method. Bioelectrochem. Bioenerg. 46, 105-114. *Explain the theory and practical application of the research behind the above patent*. http://dx.doi.org/10.1016/S0302-4598(98)00132-9

e) Yardley, J.E., Todd, R., Nicholson, D.J., Barrett, J., Kell, D.B. and Davey, C.L. (2000) Correction of the Influence of Baseline Artefacts and Electrode Polarisation on Dielectric Spectra. Bioelectrochem. 51, 53-65. *Develops an integrated method for addressing both electrode polarisation and electronic distortion of the baseline at different frequencies allowing a more robust estimation of biomass concentration from the dielectric signal.* http://dx.doi.org/10.1016/S0302-4598(99)00069-0

3.2. Patent:

Davey and Kell (Priority date 1997) Capacitance measurement of a dielectric medium. WEP1018025 / US6496020 / WO1999017124A1. *Enables design of instrument electronics that permit automated removal of electrode polarisation from biomass signals. Electrode polarisation is*

a significant source of interference in capacitance-based biomass measurement and this research improves sensitivity of the technique

3.3 Books:

a) Davey, C.L. (1993) The Biomass Monitor Source Book. Aber Instruments Ltd (Science Park, Aberystwyth). ISBN 0 9521019 0 4. *Explains operation of the instruments supplied by Aber Instruments – commissioned by Aber Instruments in response to customer demand.*

b) Davey, C.L. (1993) The Theory of the β -Dielectric Dispersion and its use in the Estimation of Cellular Biomass. Aber Instruments Ltd (Science Park, Aberystwyth). ISBN 0 9521019 1 2. *Explains theory underpinning the instruments supplied by Aber Instruments – commissioned by Aber Instruments in response to customer demand.*

4. Details of the impact

BEAA research has had continuing economic and commercial impact throughout the REF period locally on Aber Instruments and nationally and internationally on their distributors, customers and product consumers by changing the methods used by practitioners in breweries and biotechnology companies worldwide. Patenting of BEAA intellectual property has continuing impact by protecting the market for Aber Instruments (see section 3). The company has established its independent viability, increased its workforce and commercialised a range of high specification on-line instruments with worldwide patent cover, including the Futura, which largely replaced the Biomass Monitor in 2009 for biotechnology applications, and the development and improvement of the Yeast Monitor for use in breweries [5.1].

Aber Instruments' Engineering Director has confirmed [5.2] that the company "continually works with Dr Hazel Davey and her research team in an effort to understand cellular processes further and pass that knowledge on to the marketplace". He has confirmed that Dr Hazel Davey's research has impact through "her presentation at Aber Instruments' Distributors Conference in May 2013 and through other avenues such as papers, websites and posters" and that "this helps Aber Instruments to maintain its presence in the marketplace and to promote its products." He goes on to state that "without this on-going work Aber Instruments would not be able to maintain its profits" and that "Dr Hazel Davey's expertise on cell vitality and the instruments that measure it is extremely useful to Aber Instruments for exploring new market potential".

The impact of BEAA research has significantly raised the profile of Aber Instruments technology. This has resulted in it being considered a robust and verified method and has led to a change in practitioner approach to monitoring and decision making in a wide variety of processes that involve cellular material. Aber Instruments launched its third major generation of products, the Futura range, in 2009 [5.1]. The Futura is a new generation of compact, lightweight biomass monitoring instrumentation that can be used on conventional as well as disposable bioreactors. Research conducted at the university has directly contributed to these developments by providing methods to reduce and correct for interference in the biomass signals. Aber Instruments' Sales and Marketing Director has confirmed that BEAA research has led to "an extensive list of key publications that are often referred to by customers of Aber Instruments" and that the two books written by Dr Christopher Davey (see section 3) continue to be "the standard references used by practitioners in industries using Aber Instruments technology" [5.3].

In addition to the local impact on Aber Instruments, the impact of BEAA research is much further-reaching as over 1000 systems based on the technology that resulted from this research have been supplied worldwide [5.1] and these are used in a wide variety of applications and sectors. Worldwide, industries adopting Aber Instruments radio-frequency impedance technology for fermentation and cell culture monitoring include breweries, major biotechnology companies and increasingly biofuel plants. Thus, it can be seen that, via Aber Instruments, the research conducted by a team of scientists within AU is directly impacting on companies and end users worldwide. The technology developed with input from AU staff is used for research, process development and manufacturing by the world's leading biotechnology companies such as Genetech, Novartis, Novo,

Biogen Idec, GlaxoSmithkline, Centocor, Sandoz, Eli Lilly and Genzyme [5.3,5.4]. The Yeast Monitor is now established as the standard for on-line yeast concentration measurement in large breweries including AB Inbev, SAB Miller, Coors, Diageo, Heineken, Suntory, Kirin, Asia-Pacific and San Miguel [5.1,5.3,5.4]. Aber Instruments' Engineering Director has confirmed that these customers run trials prior to purchasing biomass monitoring systems to determine cost-benefit analyses and that the "research, knowledge and understanding in this area lead to cost-benefit analyses which show a typical saving in the process of 10%" [5.2].

In the brewery industry, the approach to viability measurement developed at AU is used mainly for on-line control of the rate and total amount of yeast added to fermentation vessels to convert wort into beer. It is also used for off-line measurements of yeast quality to decide the number of times yeast can be recycled from one batch to the next. This is important as recycling is quicker and reduces propagation and disposal costs. Aber Instruments' Sales and Marketing Director has confirmed that on-line pitching control alone has the following on-going benefits for breweries adopting this technology [5.3]:

1. Improving fermentation efficiency leads to cost savings of €158,184 per million hl of beer.
2. A reduction in labour costs of >€40,000 per brewery, per annum as the on line, automated monitoring of yeast quality and concentration reduces the need for technical staff.
3. Outlay on additional fermentation vessels is reduced through improving identification of brew completion via on-line, real-time monitoring. This allows the fermented beverage to be processed more quickly; for example, cutting the brew time from 5 days to 4 increases the production of the brewery by 20% without the need for financial outlay on an extra fermentation vessel. With each fermentation vessel and associated instrumentation system costing approximately €700,000 this is an important cost saving.
4. Better consistency of product flavour; this is especially important for global brands where poor consistency leads to customer dissatisfaction and loss of market share.

Locally, Aber Instruments is benefitting from BEAA research through growth of the company workforce and profits and opportunities for accessing further investment in research. The impact of BEAA research has also significantly raised the profile of Aber Instruments technology, confirming its status as a robust and verified method and this in turn has led to a change in practitioner approach to monitoring and decision making in a wide variety of processes that involve cellular material. Worldwide, industries adopting Aber Instruments radio-frequency impedance technology for fermentation and cell culture monitoring include breweries and major biotechnology companies. Thus, it can be seen that, via Aber Instruments, the research conducted by a team of scientists within BEAA is directly impacting on companies and end users worldwide.

5. Sources to corroborate the impact

5.1. Aber Instruments company profile http://www.aber-instruments.co.uk/about-us?mr=789
5.2. Statement from Engineering Director, Aber Instruments [Letter]
5.3. Statement from Sales and Marketing Director, Aber Instruments [Letter]
5.4. Economic Impact of the Institute of Biological, Environmental and Rural Sciences: Research and Operational Activities DTZ Report 2009. Available at http://www.aber.ac.uk/en/media/departmental/ibers/pdf/DTZ_report.pdf