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| Project no: | STRP 016833 |
| Project acronym: | SNiP2CHIP |
| Project title: | Development of a complete integrated SNP analysis system |
| Instrument: | STREP |
| Thematic Priority: | Sixth Framework Programme Priority 3: Nanotechnologies And Nanosciences, Knowledge-Based Multifunctional Materials, And New Production Processes And Devices |

**Publishable Final Activity Report**

Period covered: from 01/02/2006 to 31/03/2009

Start date of project: 01-02-2006

Duration: 38 Months

Project coordinator name: Dr. Paul Galvin

Organisation name of lead contractor for this deliverable: Tyndall

## Project Objectives

The requirement to screen for known SNPs (single nucleotide polymorphisms) has become one of the key challenges to be addressed to enable the exploitation of the human genome sequence, where approximately 3 million SNPs are responsible for all of the variation within the human population. Several methods and technologies are currently available for detection of SNPs, but there is a still a clear need for single platform which can deliver 100% accuracy, in a low cost, versatile, and easy-to-use integrated system. The SNIP2CHIP project was focused on the development of integrated SNP detection platforms to include modules for DNA extraction and purification from biological samples, DNA amplification, genetic characterisation (including SNP detection), signal transduction, interpretation and data analysis. The clinical challenge addressed by the project, is the need to be able to provide clinicians with immediately actionable data based on near-patient diagnosis of infectious diseases or pharmacogenetic testing.

Therefore the overall objective of the SNiP2CHIP project was to develop an integrated system for genetic analysis, capable of providing genotypic data directly from a biological sample. The research was based on the vision to enable near-patient genetic analysis (e.g. in a physician / GP surgery), by integrating DNA extraction, amplification and analysis modules onto a single miniaturised system with an easy-to-use interface for unambiguous genotype interpretation. Central to this vision is the requirement for a system that delivers genotypic data with very high accuracy at low cost, such that it provides a novel affordable technology solution for near-patient genetic analysis.

The key technical objectives of the project were as follows:

1. Development of a novel miniaturised DNA extraction and purification module.
2. Development of a novel PCR-on-a-chip module based electrowetting actuation.
3. Development of novel DNA analysis modules based on optical and magnetic sensors for SNP detection.
4. Development of an easy to use system control module with a graphic user interface.
5. Integration of the different modules together within a functional prototype system, which will be validated based on CFTR SNP detection.

## Contractors involved

| Participant name | **Participant short name** | **Country** |
|---|---|---|
| Tyndall National Institute<br>Project Co-ordinator: Dr. Paul Galvin, Tyndall National Institute, "Lee Maltings",University College, Cork, Ireland.<br>Tel: +353 21 4904030; Fax: +353 21 4270271<br>E-mail: paul.galvin@tyndall.ie | TYNDALL | Ireland |
| Commissariat à l'Energie Atomique | CEA-LETI | France |
| Instituto De Engenharia De Sistemas E Computadores Microsistemas e Nanotecnologias | INESC-MN | Portugal |
| Centro De Investigacao Em Genetica Molecular Humana, Fundaçao Da Faculdade De Ciencias Da Universidade De Lisboa | FFCUL | Portugal |
| Czech Natl. Center for Diagnosis & Treatment of CF | UCPRA.2SM.CFC | Czech Republic |
| National Centre For Medical Genetics, National University Of Ireland | NUID/UCD | Ireland |
| Asper Biotech | Asper Biotech | Estonia |

**Work performed**

Integration of different modules was implemented using a transport mechanism based on electrowetting actuation (EWOD). Two alternative SNP detection platforms were developed based on optical and magnetic sensing respectively. The different modules developed were designed to be compatible with integration as a single automated functional prototype system with a simplified GUI interface, based on end-user specifications. The systems development focused on ultimate delivery of a prototype system customised for low to medium throughput, low cost, point-of-care applications, with emphasis on providing very rapid and accurate results. Within the project, screening of SNPs in the CFTR gene associated with cystic fibrosis was evaluated. CF patient samples were used to verify the accuracy and reproducibility of the system in a clinical diagnostics laboratory.

**End Results**

The main goal of the research is the development of a novel integrated mutation detection system, in which each of its modules have innovative solutions for DNA extraction, amplification, and DNA microarray based detection with either optical or magnetic detection. The project has demonstrated working modules for all of the required building blocks, and integration was demonstrated with some of the modules as a proof of concept for a complete integrated system.

- The integrated genetic analysis system architecture provides the basis for a novel solution for near-patient genetic analysis. However, the different modules which were developed within the system are also highly innovative.
- The DNA extraction module comprises a novel microsystem for automated DNA extraction using established paramagnetic bead-based protocols for DNA extraction and purification in a lab-on-chip format.
- The PCR amplification module comprises a novel microsystem for PCR amplification using channel-free transport of microdroplet reactions across temperature zones using electrowetting actuation (EWOD).
- The magnetic sensor based DNA microarray platform is a novel, rapid and potentially low cost solution for highly parallel SNP detection
- The optical flow-through platform provides a rapid, low-cost solution for low to medium throughput SNP detection

The next steps to facilitate the commercialisation of the SNIP2CHIP integrated system involves optimising the device and system architectures to facilitate high volume manufacturing while ensuring the level of accuracy and reproducibility required for clinical diagnostics is maintained. Due to the modular nature of the system, it is envisaged that commercialisation may involve taking the best solutions which have been generated within the SNIP2CHIP project and combining them with externally sourced optimised modules, to ensure that the best possible system can be available within the shortest timeframe. The key challenge remaining towards product development from the SNIP2CHIP outputs, is to complete the integration of the system as a low cost solution, both for the instrument (e.g. less than €5000) and the disposable cartridge for each patient sample (e.g. less than €10).

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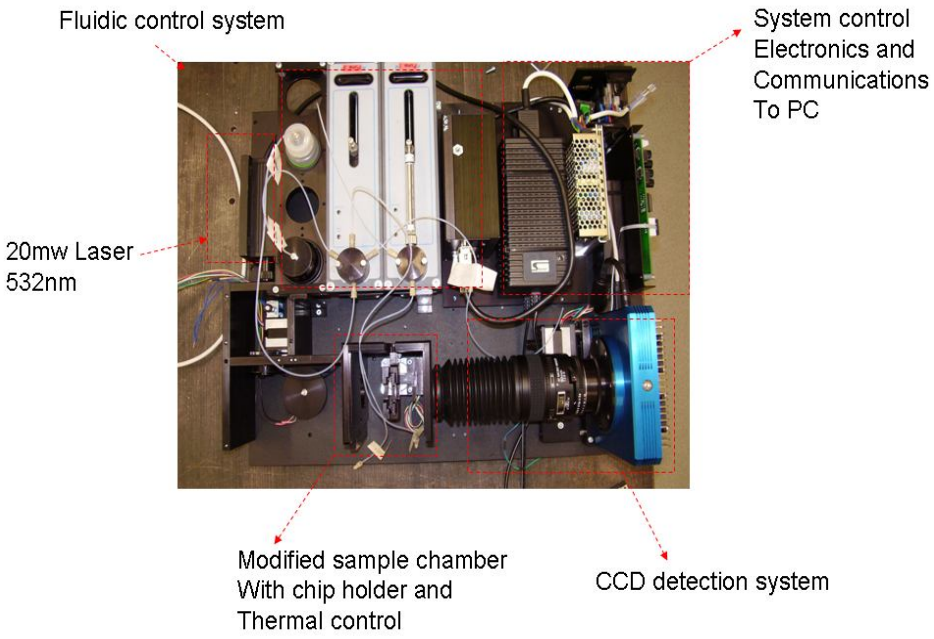


**Figure 1: Integrated optofluidic mutation detection system including fluidic control pumps and valves, thermal control device, and holder for the microfluidic cartridge**

**Impact**

In the post genomic era, together with many technical and scientific advances of the last twenty years of the 20th century, there has been a dramatic increase in the demand and provision of genetic testing both in the EU and also in rest the developed world. The use of genetic tests for humans – for diagnostic, confirmatory and predictive purposes – is expanding in all European countries. Laboratories face a proliferation of demand for molecular genetic testing for a wider range of diseases. Following the completion of the first draft of the human genome sequence, the results have been translated into medical applications including diagnostics and therapy and promote new concepts in predictive medicine. Several hundred hereditary diseases have already been assigned to specific genes and their mutations. The number of diseases for which genetic tests are available will continue to increase in the coming years. Additionally, genetic testing for predisposition to numerous human malignancies and to multifactorial diseases is being considered, opening up the possibility of making personalized or targeted disease prevention a new paradigm of medicine. Pharmacogenetics based on common genetic variants influencing effectiveness of pharmaceuticals and aimed at developing and prescribing the right medicine for the right patient signifies another line of future possible applications of SNP detection in medicine.

The core originality of this technology will be the integration of DNA sample preparation and multiple SNP analysis in a single automated system. Despite the growing demand for accurate SNP data for clinical diagnosis of monogenic diseases, no products currently exist which enables multiple SNPs to be screened directly from a blood sample within a fully integrated system.

The SNiP2CHIP project consortium included examples of initial end-users of the technology i.e. clinical laboratories in both the design specification and validation of the system, to ensure that the system developed will be appropriate for the requirements of the clinical diagnostics market. The inclusion of Asper Biotech as an SME in the consortium, helped guide the development of clear exploitation strategies for outputs of the research.

**SNiP2CHIP** **Project No: STRP 016833**

The SNiP2CHIP project targeted the development of a low cost easy-to-use system accessible to the non-specialist. By ensuring that both operation of, and data interpretation from the system is compatible with the non-specialist, and that the system could deliver a genotype within 1-2 hours of collecting the sample, a market among general practitioners and physicians is easily envisaged, especially in the case of the larger medicentres with greater resources.

The availability of an automated and rapid integrated system for genetic analysis of patient samples has the potential to form the basis of a "disruptive technology", which could significantly improve healthcare delivery. By affording the possibility for near-realtime (e.g. <60 minutes) analysis of patient DNA for specific traits, on-site at their local medical clinic / general practitioner, the duration of anxiety of the patient can be greatly reduced. This technology will simplify the delivery of a universal high standard of genotyping for clinical diagnosis, which currently requires specialist clinicians and a range of expensive equipment usually found in only one or a few laboratories in each state. The integration of the processes into a single system will eliminate a significant source of error, due to mistyping of patient samples caused by either technical or clerical errors (e.g. mixing of samples during the different steps of the analysis process). In the long-term, SNP screening technology such as the SNiP2CHIP system, will be essential for personalised medicine to enable selection of the drugs and dosages appropriate to each patient.

**Dissemination and use**

The project has developed innovative solutions for DNA extraction, amplification and SNP detection (based on optical and magnetic sensing platforms), and the development of a novel integrated system which enables complete processing of all molecular steps necessary to deliver genotype outputs directly from a finger stick blood biopsy. Various potential innovations have been explored with one IDF filed on the optical sensor platform. However, following an in-depth Patent review, similar IP preceding the snip2chip IDF file date was discovered which greatly eroded the potential patentability of the invention.

**Overview table**

| **Exploitable Knowledge** (description) | **Exploitable product(s) or measure(s)** | **Sector(s)** of application | Timetable for commercial use | Patents or other IPR protection | **Owner** & Other Partner(s) involved |
|---|---|---|---|---|---|
| 1. New optical SNP detection method and technology | Genotyping instrument | 1. Medical diagnostics<br>2. Drug discovery | 2012-2015 | A joint invention disclosure was prepared in 2008 relating to a novel concept for enabling optica SNP analysis, and submitted to the TTO office in Tyndall. | Tyndall and Asper |
| 2. New magnetic sensor platform for genetic analysis | Rapid genetic analysis instrument | Medical diagnostics | 2012-2015 | A joint patent between INESC-MN and Tyndall has been filed | INESC-MN & Tyndall |

| **Exploitable Knowledge** (description) | **Exploitable product(s) or measure(s)** | **Sector(s)** of application | Timetable for commercial use | Patents or other IPR protection | **Owner** & Other Partner(s) involved |
|---|---|---|---|---|---|
| | | | | in US and Europe | |
| 3. Market gap analysis for integrated systems for mutation testing | Technical specifications | 1. Medical diagnostics 2. Clinical trials | 2011-2015 | NA | Snip2chip consortium |
| 4. Protocols for multiplexed DNA amplification of CFTR genes | Proprietary knowhow | Medical diagnostics | 2011-2015 | NA | Snip2chip consortium |
| 5. Knowhow for integration of heterogeneous devices into biochips | Proprietary knowhow | Medical diagnostics | 2011-2015 | NA | Snip2chip consortium |

## Dissemination

In the interest of ensuring publicity of research ongoing within the project, a number of disclosures were made using information that would not compromise any potential IP from the project as follows.

**Overview table**

| **Planned /actual Dates** | **Type** | **Type of audience** | **Countries addressed** | **Size of audience** | **Partner responsible /involved** |
|---|---|---|---|---|---|
| Since March 2006 | Project web-site | All | International | Not quantified | Tyndall |
| June 2009 | Presentation: *Development Of an Integrated EWOD Based POC system for Genetic Analysis.* D.Brennan et al. NanoBioEurope 09 Grenoble June 16- 18th | Scientific | International | 150 | Tyndall & all |
| June 2009 | Presentation: *Development Of an Integrated EWOD Based POC system for Genetic Analysis.* D.Brennan et al. EuroNanoForum 2009, Prague 2-5 June. | Scientific | International | 150 | Tyndall & all |
| June 2009 | Presentation: *Development Of an Integrated EWOD Based POC system for Genetic Analysis.* D.Brennan et al. Translational Health | Scientific | Irish | 50 | Tyndall |

| Planned /actual Dates | Type | Type of audience | Countries addressed | Size of audience | Partner responsible /involved |
|---|---|---|---|---|---|
| | Research Conference 24 June 2009 in Cork, Ireland. | | | | |
| Oct 2008 | Presentation: 22nd Annual North American Cystic Fibrosis Conference (NACFC) in Orlando, Florida on October 23rd-25th. L Clarke | Scientific | International | 300 | FFCUL |
| Aug 19th and 20th 2008 | Presentation Enabling Point-of-Care Diagnostics Conference in Washington. M Dobson | Scientific | International | 300 | NUID/UCD |
| Aug 21st & 22nd 2008 | Presentation The Future of Cancer Diagnostics - Next Generation Molecular Technologies M Dobson | Scientific | International | 300 | NUID/UCD |

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Several manuscripts are currently being prepared for publication in high quality international peer-reviewed journals, while ensuring that all IP issues have been fully clarified. These will include publications on each of the modules for DNA extraction, amplification and both optical and magnetic detection. Additionally, publications are also anticipated resulting from the development of SNP detection platforms.

TV Footage was taken of some of the snip2chip devices system for an Irish scientific documentary "The Investigators", which may be broadcasted in a future series.

A video was made of the research from the project as part of a scientific outreach activity, "Your Science, Your Say", to get public reaction to the concept of the genetic analysis system and to help identify potential issues or concerns that the public may have relating to the development of the technology.