Strategy paper
Pathogenomics
ERA- NETWork

Pathogenomics, Innovation and Public health

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This document is meant to be a discussion paper and is therefore open for changes and suggestions. This work was financed by the European Community through the ERA-NET "Pathogenomics" programme.

Executive summary

The field of anti-infective drugs is facing a crisis due to three major factors: (a) pathogens are constantly developing resistance to existing drugs (especially at the hospital setting); (b) the development of new antibacterial agents is nearing a standstill (new antibacterial agents constitute merely 6 of 506 drugs disclosed in the developmental programs of the largest pharmaceutical and biotech companies in the U.S.); (c) a dearth of reliable diagnostic and monitoring tools for infectious disease complicates treatment and diminishes its efficiency.

Despite the ever growing need for new antimicrobial agents, a number of factors make their development economically unattractive. First, health authorities agree on the need to limit the use of broad spectrum antimicrobials in order to minimize the selective pressure driving resistance. Second, the products are almost certainly short-lived, since resistance to the product is likely to rise with time. Third, aging of the Western population has shifted drug discovery efforts towards agents that treat chronic medical conditions. Finally, newly developed agents face a strong competition, due to the large number of antimicrobials already approved.

In addition, the field suffers from four major setbacks: lack of standardization, insufficient collaborations, shortage in publicly available, comprehensive databases and lack of legislative groundwork. High throughput methods, such as genomics and proteomics, while helping to propel the field forward, accentuate these setbacks by providing overwhelming amounts of data that have yet to be properly handled.

The genomics era initiated large-scale identification and validation of biomarkers and drug targets. The vast amount of data generated should be deposited in open access databases that will enhance the development of fast, serological or molecular tests, with the possibility of quantification. Significant efforts should be invested in creating and optimizing *in silico* tools that could capitalize on this wealth of information.

Although the "-omics" methodologies did not, perhaps, propel the anti-infective field at the expected pace, they nevertheless play an important role in target detection and validation, efficacy and toxicity monitoring, determining the mode of action of a molecule and clinical trial optimization. In fact, due to genomics-based improvements in drug discovery efficiency,

bacteriology is one of the rare disciplines where pre-clinical data can predict, to a certain extent, efficacy in clinical development. This means that there are fewer candidate antibacterial compounds entering clinical development than in other disciplines. Unfortunately, this is often perceived as a weakness, but in fact, even more efforts should be put into the establishment of "tailored" compound collections that have a potentially increased rate of success.

While such compounds historically target various cellular functions of the pathogen itself, virulence factors should also be considered as potential targets, since targeting pathogenic virulence rather than survival may lead to a reduced selection pressure and, thereby, fewer drug-resistant mutations.

As with any applied science, innovative anti-infectives and diagnostic and monitoring tools will evolve only from the collaborative efforts of academia, biotech and big pharma. Technology Transfer Offices (TTOs) are responsible for the first steps of prioritizing and patenting new inventions originating in the academia, and cultivating interactions with the industry. TTOs in Europe seem to be less productive than their counterparts in the U.S., and optimizing their performance is one way to enhance commercialization of basic research inventions.

Ongoing attempts to establish a Community Patent in Europe also hinder commercialization of inventions, since patenting across several or all of the EU Member States is much more complex and costly compared to the U.S. Bureaucracy, costs and lengthy processing create barriers to patenting and affect the innovation performance in Europe.

On the other hand, over-patenting could also take its toll. Especially in the era of functional genomics and the emerging field of systems biology, patenting could lead to appropriation of whole biological pathways and hinder further research. Therefore, it is of uttermost importance that a political and legal framework should be setup to ensure that research and patenting remain accessible to all research groups. Political support is also warranted in the form of initiatives and economic incentives as well as updated legislation that will address the new reality being forged by the various "-omics" methodologies.

The following points summarize the caveats and needs of the anti-infectives field in the context of pathogenomics:

1. A substantial amount of time, efforts and resources seem to be consumed due to lack of standardization between labs. Formulation of guidelines relating to research tools and nomenclature could aid in communications between labs and streamline research.
2. Networking networking…connecting academia, the industry and legislators across the EU.
3. There is a shortage of biomarkers that could serve as diagnostic tools, monitor the course of the disease or predict its likely outcome. Genomics and related techniques are extremely efficient for identifying such markers.
4. Legislation is falling behind scientific research on several fronts, namely the use of biological markers, patenting, financing and drug approval procedures.
5. The novel high throughput research techniques generate an overwhelming amount of data regarding pathogens, targets for drug intervention, candidate compounds and biomarkers. Open access databases are crucial for efficient research, and as such should take high priority, along with computational tools allowing efficient data mining.

I. Introduction

This paper aims to outline the possibilities of commercializing the fruits of genomic research on pathogenic microorganisms (bacteria and fungi), as well as to highlight existing barriers that impede an efficient implementation process. We delineate the multi-step process of transforming a fundamental finding into a tool for the benefit of society, beginning with the scientists, and continuing with technology transfer officers, biotech companies, industry, health care professionals and policy makers. This development process, be it for a drug, a diagnostic tool or a vaccine, should, of course, remain highly attuned to the patients' needs and to the particular disease it aims to treat or diagnose.

First, we will delineate the economic, legal and social background of the technology transfer process from the public to the private sector, an important force propelling innovation. Thereafter, more specific issues related to the "-omics" research in infectious disease will be discussed, with special emphasis on anti-infective agents, diagnostics, monitoring tools and vaccine development.

Innovation, genomics and infectious diseases

Despite extensive public research activity (319,777 PubMed publications on "infection and bacteria"), and, consequently, elaborate knowledge about causative pathogens and how to combat them, there is a certain disinterest in the development of anti-infectives. Indeed, new antibacterial agents constitute merely 6 of 506 drugs disclosed in the developmental programs of the largest pharmaceutical and biotech companies in the US (Internet listings and 2002 annual reports, Spellberg *et al.*, 2004). Despite a potential antibacterial market of approximately 25 –28 billion $ (Bax, 2001), several pharmaceutical companies have indicated that they are curtailing anti-infective research programs.

A number of factors make the development of antimicrobial agents less economically attractive than other drug classes. First, health authorities agree on the need to limit the use of broad spectrum antimicrobials, thus minimizing the selective pressure driving resistance. Second, the products are almost certainly short-lived, since resistance to the product is likely to rise with time. Third, aging of the Western population has shifted drug discovery efforts towards agents that treat chronic medical conditions. Finally, newly developed agents face a strong competition, due to the large number of antimicrobials already approved.

Yet, although they are not economically attractive, anti-infectives are vital for maintaining public health. Indeed, infectious agents are still the second most important cause of death worldwide (WHO, 2001). HIV/AIDS is on the rise and associated opportunistic pathogens (e.g. Candida) are significant causes of mortality. Tuberculosis (TB) is making a come-back, killing roughly two million people every year (MSF publication 2005), and pneumonia has become the third most frequent reason for hospitalisation, after births and heart diseases (www.reutershealth.com).

Despite the large variety of existing anti-infectives, multidrug resistant (MDR) pathogens such as *Mycobacterium tuberculosis* are emerging. Especially in Eastern Europe, where nearly half of all TB cases resist at least one first line drug. For example, in Latvia there is a 20% prevalence of poly-drug resistance and 12% of MDR-TB (WHO/IUATDL survey, 2000). Resistant *Staphylococcus* and *Enterococcus* strains are a rising threat in developed countries, not only in intensive care units but also in unexpected community settings such as sport clubs (Appelbaum, 2006). Clearly, the concomitant rise in the number of infections and in drug resistance shows no sign of abating.

In addition to new treatments, reliable diagnostic tools for the rapid detection of important pathogens (e.g. TB in HIV positive patients) are painfully lacking and a standardized methodology to assess resistance to second line drugs has not yet been formulated. It is estimated that only a minority of all living bacteria are characterised. Genomics and related molecular techniques are crucial for the study of pathogens that cannot be cultivated. Therefore, it is important to allocate resources for the development of genome-based molecular methods, especially since some of these unidentified pathogens might be linked to unexplained diseases. In addition, better models, (e.g. humanised mice, representative cellular models and more), representing the natural environment and host interactions of the studied pathogens should be sought for.

Unfortunately, new technologies based on functional genomics have not yet created the expected boost for drug discovery. Nevertheless, they play an important role in target detection and validation, efficacy and toxicity monitoring, determining the mode of action of a molecule and clinical trial optimisation. Genomics and related methodologies are also expected to instigate disease diagnostics and monitoring by identifying novel biomarkers.

Such advances could prove especially useful for point of care applications (fast and easy tools) and for the detection of populations at risk.

II. Global R&D - Background

Product development and Innovation is a multi-step process with multiple contributors. Figure 1 illustrates the different actors involved in the development process of drugs, monitoring tools and vaccines for infectious diseases. Figure 2 outlines the FDA's model for the stages of drug development.

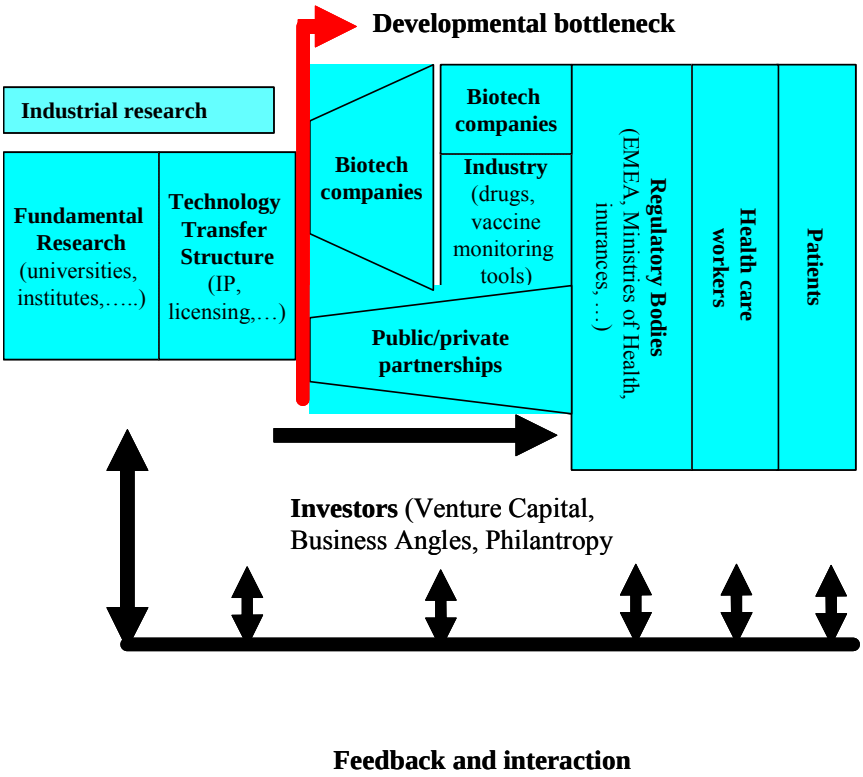

Figure 1: The different actors in the development process of health care applications (drugs, monitoring tools and vaccines).

An invention is usually transferred, after patenting, from the academic laboratory to the private sector via a technology transfer office. A licensing agreement usually transfers the rights for further development to the private sector.

In the past, big pharmaceutical companies dealt directly with the academic level in search for promising inventions. At the end of the last century, however, biotechnology companies took over the role of the intermediary between invention and application. But in 2001, the economic climate changed and Venture Capitalists (VC) became reluctant to put their faith, and their money, in novel drugs that have the potential to become blockbusters 10-15 years down the road, preferring, instead, to enter the scene at a later, much less risky stage. This created a development gap between basic research and its possible applications (Figure 3). Today's challenge is to bridge this gap and so ensure the continuity between invention and product.

Another factor undermining the involvement of the private sector in anti-infective research is the mergers of big pharma companies, which tended to increase over the last 15 years. These mergers are evidently driven by economic considerations, since they enable to cut down production, commercialisation and R&D costs. One of the consequences, however, is that certain research fields, such as anti-bacterials and anti-fungals, may be aborted while others remain, such as the lucrative anti-virals with a worldwide market and long term prospects (Overbye & Barett, 2005).

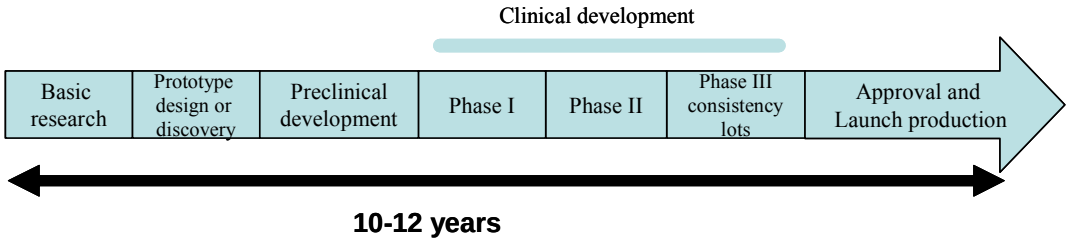

Figure 2: FDA's Critical Path for medical product development model (adapted fromPhillips, Van Bebber and Issa 2006).

Phase I: First human trials. Assess safety in healthy human volunteers (10-20 people)
Phase II: Study to evaluate dosage, efficiency and side effects (20-100 people)
Phase III: Study to evaluate efficiency compared to existing products and adverse reactions to longer term use (100-1000-5000 people)

Adapted from DNDi, www. Dndi.org

Over the last ten years a new type of development structure has appeared, the Public Private Partnerships (PPPs), which link big pharma and biotech with academia, non-governmental organisations and multilateral groups such as the World Health Organisation (WHO). Today there are nearly 100 PPPs with a combined total budget of 1 billion $ (Cohen, 2006). Eighty per cent of them are funded by non-profit organizations (Rockefeller Foundation, Bill and Melinda Gates Foundation, etc.) and they account for nearly 75% of R&D projects focusing on drugs to treat neglected diseases (Cohen, 2006). Companies that join PPPs have little prospect of profiting on the drugs they develop, but also a relatively limited financial risk because their partners typically pay for the most expensive part of the process, namely large-scale clinical trials. This "no profit-no loss" business model offers big pharma various benefits, including a good public image, an introduction to developing-country markets and contacts with researchers.

Once a product has passed the clinical development stage, it needs to be approved by regulatory bodies (FDA in the US, EMEA in Europe). In Europe, regulatory approval leads to implementation by the various ministries of health and to reimbursement by social security systems. The companies introduce their product through marketing initiatives and finally the clinicians prescribe the drugs, monitoring tools or vaccines to the patients. The clinicians are also the first to evaluate the long term impact of those products on treating, monitoring and preventing disease.

Fig3: Lack of funding is creating a financial desert at early stages of development.

Important financial gap at the early development stage

II.a. Steering universities towards Innovation

The Bayh-Dole Act (1980, US law), marked a world-wide change in university-industry relations. The Act provides a policy framework to encourage universities and other non-profit organisations to patent and commercialise inventions and new technologies by collaborating with commercial enterprises. The aim of the Bayh-Dole Act was to translate academic research into jobs and financial returns, giving the universities a new role within the national innovation system. This rationale has inspired subsequent initiatives elsewhere in the industrialised world (mainly in Europe and Japan), although the university is not always the owner of the Intellectual Property (IP) rights (Table 1). Overall, there has been a noticeable international effort towards engaging university scientists in "patentable" research and towards involving their respective institutions in licensing IP based upon publicly funded research results.

In order to facilitate this process, it should be as streamlined as possible so that researchers are not discouraged by the administrative load and the unfamiliar legal and business aspects. Also, researchers should be duly informed about royalty-sharing formulas in their institution, since universities and institutions in Europe and North America have adopted a wide range of different policies on patent ownership and royalty-sharing formulas between inventors, the inventor's department, the Technology Transfer Office (TTO) and the institution itself (Table 1).

	Owner of IP rights
Finland	**Inventor (will change very soon)**
Germany	**University/institution**
France	**University/institutions**
Spain	**Institutions**
UK	**Increasingly from funding agencies to universities**
Italy	**Researcher**

Table 1: Differences across Europe concerning the ownership of IP rights

What is a patent ?

A patent is an exclusive right granted by the State for an invention that is new, involves an inventive step and is capable of industrial application.
It gives its owner the exclusive right to all applications related to it.
A patent is granted by the national patent office of a country or a regional patent office for a group of countries. It is valid for a limited period of time, generally 20 years from the date of filing of the application. A patent is a territorial right, limited to the geographical boundary of the relevant country or region.

Source: http://www.wipo.int

Box 1: What is a patent

II.b. Patenting strategies

There are two main patenting strategies. According to the first one, only the most promising inventions with a clear market potential will be patented. This can be considered as a risky approach, mainly because the commercial potential of a novel technology may be difficult to gauge during the early stages.

Output	Mean US	Mean Europe
Number of patents filed	35.8	6.2
Number of patents issued	16.8	5.8
Number of active licensing contracts	120.2	17.1
Revenue from licences (1000€)	10,173	507
Number of spin-offs	2.1	1.6

The number of patents filed or issued by European Technology Transfer Offices (TTOs) has been much lower than those for the US. The average number of active licensing contracts amounted to about 120 in the US and only 17 in the EU. Even when considering that European and US patent systems are not fully comparable, this suggest a lower level of activity of European TTOs. The number of spin-offs is relatively similar. These numbers might suggest directing efforts at the EU level to make TTOs more outward oriented by improving their marketing and communication strategies towards SMEs, Industries and their own publicly funded research organisations (From: Expert group report EU, 2004).

Table 2: Comparison of technology transfer performance indicators in the US and Europe, 2002

According to a second strategy, all or most of the inventions are evaluated for patenting. Casting a wide patenting net ensures catching the “big fish”, which will give a considerable return on investment, while generating many modest revenues in the meanwhile.

Still, it might be counterproductive to seek patent protection on nearly everything. Privatization and commercialization of biomedical research must be carefully deployed to sustain both upstream research and downstream product development. On the one hand, commercializing biomedical inventions is crucial for financial growth, and on the other hand, IP issues should not constitute a barrier to the research process itself. Indeed, patents tend to have an inhibiting effect on research and related clinical practice (see also Intellectual

property, systems biology and multidisciplinary approaches). If research is to be allowed to flourish freely, precompetitive findings should be shared as much possible, at least on the academic level (as done in the Human Genome Project), although a precise definition of "precompetitive" is still lacking. The implications of patenting on public health should also be considered, and are continuously discussed in the scientific community. For example, in the past, misuse of patent rights lead to prohibitively expensive tests for life-threatening diseases (e.g. BRCA1 and 2 genes related to breast cancer). At issue is also whether there is an obligation to the public to ensure that clinical and research access to valuable discoveries is not unduly restricted.

The BRCA story

In 1994, the research group of Mark Skolnick at the University of Utah identified BRCA1 as a gene underlying hereditary breast cancer. Skolnick and the University of Utah were granted a U.S. patent for the gene sequence of BRCA1. They licensed the exclusive rights to Myriad Genetics, a biopharmaceutical company founded in 1991 by Mark Skolnick and others.
Soon afterwards a second gene also responsible for hereditary breast cancer was discovered (BRCA2), this time by two groups: Myriad Genetics and the Institute for Cancer Research in the United Kingdom. Certain mutations in the BRCA1 or BRCA2 genes disrupt the regulation of growth of mammary cells - a critical step on the path to tumor formation. Myriad Genetics, in essence, had a monopoly over diagnostic testing for BRCA1 and 2 familial breast cancer in the United States.
Myriad Genetics began enforcing its patent claims against certain universities. To curb criticism from the academic community, in 2000, Myriad Genetics negotiated an agreement with NIH so that NIH-funded researchers would receive a discount on Myriad's BRAC analysis test as long as the test was used for research purposes. In exchange, Myriad Genetics would have access to resulting research data.
Myriad Genetics also sought patent protection on BRCA1 and BRCA2 in the EU. The European patents granted to Myriad Genetics covered any method for diagnosing a predisposition for breast and/or ovarian cancer, 34 specific mutations, the BRCA1 gene itself, the corresponding protein, therapeutic applications of the BRCA1 gene, diagnostic kits and materials and methods used to isolate and detect BRCA2. In 2001, 2002 and 2003 researchers challenged Myriad's patent on BRCA1 and 2. Most of the complaints fell under the categories of failure to show novelty, inventive step, industrial application and disclosure. The patent on BRCA2 was finally granted to the Cancer Research UK, who filed it first, and who plans to allow free public access to its patented gene sequence. The sequence of BRCA1 fell into the public domain (2004) after Myriad's first patent was struck down based on errors in the original sequence and lack of an inventive step. These rulings, made effective by the European Patent Office (EPO), were considered a victory to those supporting free access to BRCA1 testing in Europe.

(From Merril & Mazza, National Research Council publication, 2006).

Box 2: The BRCA story

The ideal balance, therefore, is difficult to attain and requires good insight and considerable experience of TT-officers. Therefore, in most research institutions a Technology Transfer Office (TTO) has been created to encourage public-private collaborations and facilitate the commercialization of university research results. Technology transfer should be seen as a long

term process. It is difficult to predict when the "Big Project" will come, but when it comes the TTO should be ready to manage and defend it if necessary. An important objective and challenge in the postgenomic era, characterized by multidisciplinary and transversal research, will be to organise the legal framework such that the transfer is not aborted due to legal entanglements.

In Europe, there is an additional complication, namely the ongoing attempts to establish a Community patent. Currently, patenting is much more complex and costly for those who seek IP protection across several or all of the EU Member States, constituting a clear disadvantage in comparison to the US. Bureaucracy, costs and lengthy processing create barriers to patenting and affect the innovation performance in Europe (Charles River Associates, 2004).

II.c. Technology Transfer Offices: from public to private sector

II.c.1. Interaction with academics

Technology transfer officers are the mediators between fundamental and applied disciplines. They should actively increase awareness of researchers to the TTO, to the benefits of commercializing research and to the appropriate procedures. A successful technology transfer framework within academic institutions also relies on casual gatherings where scientists, TT-officers, clinicians and industry representatives can exchange ideas. Training and awareness raising therefore constitute an important added function of the Technology Transfer process. There is a need to stimulate communication between TT-officers, scientists, clinicians and the industry.

The technology transfer landscape is very heterogeneous across Europe (Table 3), mainly due to the fact that technology transfer is still in its infancy, and financial backing of this profession varies between countries. TTOs across Europe are differently organised (public or private) and financed, and consequently vary in size and capabilities. There is an urgent need for harmonisation of the transfer process at the European level and for the creation of a European Tech Transfer educational programme, which will enable exchange between different institutions. Furthermore, technical assistance programs, collaborative research, training, staff mobility and professional development can shape direct, long-term collaborations between university and business.

BE	DK	DE	EL	ES	FR	IE	IT	LU	NL	AT	PT	FI	SW	UK
17	31	334	22	165	209	26	93	7	20	31	20	27	58	165

Table 3: Number of Technology Transfer Offices in EU-15 by Member State in 2003 (From: Expert group report EU, 2004).

II.c.2 Interactions with the private sector

Academic merit is often evaluated by the number of publications in high ranking journals, thus favouring innovative research and new insights, and shunning, to a certain degree, clinically oriented research. Therefore, while drug discovery research efforts generally start within the academic groups, these groups tend to focus on biological activity while neglecting proof of principle or pharmaceutical drug ability. Therefore technologies emerging from university research laboratories are generally not mature for marketing: they require a significant amount of applied research and development before they become attractive to big pharma companies (Table 4).

Post-research investment is typically significantly higher than the costs of basic research. This is why patents are crucial. By providing exclusivity over the commercialization of an invention or new technology, patents provide an incentive to industries to develop that particular product. At the same time, royalties obtained from licensing a given technology can provide an incentive to the researcher and university.

Factors considered by Big Pharma when a licensing candidate:

- **Selectivity** - for one specific receptor, enzyme or ion channel
- **Potency** - *in vitro* and *in vivo*
- **Mechanism –** evidence of interaction with the target in animals
  - In a pharmacodynamic assay
  - Preferably, proof of efficiency in a validated animal model for the disease
- **Toxicology** - preliminary data
- **Bioavailability** - oral for small molecules, or other, plus a satisfactory half-life

Box 3: A good drug candidate according to the biopharma industry (adapted from MSD)

Yet, even after these initial hurdles are crossed, many worthy inventions fail to get commercialized. Contributing factors may including the actual financial/economic background and the absence of a good TTO. TTOs should promote high visibility (e.g. via websites) and a clear marketing strategy. TTOs are, so to speak, the "display window" of their mother institutes. As important as it is to have a close contact with scientists it is also important to establish a good rapport with industrial partners. Indeed, in addition to business meetings, informal contacts and the exchange of business cards are important elements which will contribute to successful negotiations and eventually to a licensing agreement.

In general, the commercialisation of research results can follow one of two main routes: a) licensing of the invention to one or more companies or b) the creation of a spin-off company that will bring it to the market. Genomics related products, software, databases, *in silico* methods and gene expression profiling, all are typically dominated by biotechnology firms. The protein structure category has been led by pharmaceutical companies, while universities have been dominant in the modified animals category (Merrill and Mazza, 2006).

In all cases, financing of the pre-clinical stage of development is difficult, particularly at the European biotech arena (Table 4) and in the field of infectious diseases. Although the biotech companies in Europe have a comparable yield to that of the US (products in clinical development/staff employed), the investment is only one third, resulting in half of the amount of products in European development pipelines. But the real question is: do we need more products or do we need more effective ones, and what should we do to succeed in delivering them to the patients who need them?

Although the development of novel drugs in general and anti-infectives in particular is problematic worldwide, especially in Europe there is a need to boost Biopharmaceutical R&D and monitoring tools, both at the biotech and the large industrial level. Therefore it is important that initiatives such as the European Associations for Bioindustries (Europabio) should be supported and promoted. It is in this context that particular problems related to the development of tools for the infectious disease field will be discussed in the next section of this paper.

Economic comparisons between the US and EU Biotech Industries		
	USA	**Europe**
Biotech companies	1830	1976
Staff employed	172,400	94,000
R&D investment	16.4 B €	6 B €
Venture Capital	2.1 B €	750M €
Debt financing	4.3 B €	1 B€
Products in clinical development	1,110	450

Table 4: Employment and financing in Biotech companies in the US and Europe

(Conference report : the EFGCP Annual Conference 2006: Retooling European Medicines Development for Leadership)

Box 4: TTO activities and functions

Some of the tasks of an Innovation department (Tech Transfer):

- Apply, explain and simplify as much as possible the most appropriate patenting procedure for the scientific partner.
- Survey the actual market needs (fields of interest of industry).
- Match basic inventions with market needs.
- Accompany the scientists during the transfer procedure, and convey the expectations of the industry at the pre-clinical level:
    1. Reach an agreement between the academic and commercial representatives on the expectations and requirements for a product prior to any further development (proof of concept: up to what level; validation of target: to what extent (in vitro, in vivo), IP status of the product, etc.).
    2. Application forms presented to the industry should be concise and informative.
    3. Standardize as much as possible "agreement formats" according to the type of product and the stage of development

General Innovation needs - summary

- Lack of communication between scientists and the industry:

*** Need for interactive meetings, congresses and communication tools**

*** Need for more tools in order to match market needs with basic inventions e.g. interactive sites with proposals from both sides.**

*** Need to inform the scientific community about IP, proof of concept and licenses issues.**

- Lack of an effective technology transfer network accessible to all academics

*** Need for a financial and legal framework at the European level, including:**

- clear agreement formats and guidelines, according to the type of invention, in order to determine the level of development needed prior to a transfer agreement.

- a European Technology Transfer educational program, which includes solutions for exchange between different European TTOs, in order to standardize European technology transfer.

- a more efficient financial, structural and strategic backbone for biotech industry in Europe. For example, shorten the time window between an application proposal and its financing (which currently may be as long as a year)

III. Impact of microbial genomics on control of infectious diseases

III.a. The "-Omics": achievements and expectations

Modern pathogen microbiology has undergone major changes during the genomic era: more than 226 whole bacterial genome sequences became available (http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?db=Genome), and novel screening technologies, proteomics, bioinformatics and new disciplines, such as systems biology, are emerging. Molecular discoveries led to new technologies enabling a high throughput study of the genome, transcriptome and proteome. These achievements enable, to a certain extent, to circumvent existing problems in pathogen culturing and the absence of adequate models, by facilitating *in situ* analysis.

In fundamental science, these novel high throughput techniques are used to acquire novel insights into the molecular mechanisms underlying pathogenicity and virulence. In applied

research, they are used in three main aspects: a) identification and validation of novel targets and biomarkers; b) development of novel screening assays to find inhibitors for these targets; and c) to determine and validate the mode of action of identified lead compounds.

The "-omics" (genomics, transcriptomics, proteomics and metabolomics) techniques contributed mainly to the first aspect. The "-Omics" brought about the era of vast amounts of data. Coupled with bioinformatics, mathematical modelling and functional molecular technologies they can provide important insights into biological systems and identify possible targets, biomarkers and mode of actions. Currently, however, these new experimental technologies are underutilized, due to a lack of scalable database systems and computational tools for target discovery (Fischer, 2005).

The challenge of modern post-genomic research, then, is to learn how to integrate knowledge from a range of these high-throughput technologies. Naturally, the promise of some of the new approaches is not yet realised, and yet they contribute to scientific progress, and will likely be more beneficial in the future.

target: A target could be an enzyme, receptor or other protein that can be modified by an external stimulus. The definition is context-dependent and can refer to the biological target of a pharmacologically active drug compound, or the receptor target of a hormone (like insulin). The terminology is that a target molecule is "hit" by a signal and its behaviour is thereby changed. The term "target" is commonly used in pharmaceutical research to describe the native protein in the body that is modified by a medicinal chemical Or identified by the diagnostic kit.

biomarker:
A biomarker is a molecule used as an indicator of a biological state or disease. for example, the presence of an antibody may indicate an infection by a specific pathogen. The biomarker may be native to the body or native to the pathogen.

Box 5: Target and biomarker definition

III.b. Anti-infective research and tools

Prior to the genomics era, the search for antibacterial agents relied primarily on screening libraries of chemical or natural components, in search for compounds that inhibit growth of the desired spectrum of bacteria. Promising drug candidates were further profiled in vivo and the molecular target was often identified after the compound had reached the market.

In modern drug discovery, where new techniques and functional genomics are playing an ever increasing role, the starting point is very often the target molecule itself. This helps to considerably reduce the pre-clinical risk (Fig 4). Indeed, the "-omics" dictate a more informed

and rational approach in target selection. For example, comparative genomic studies can give an idea on the specificity or selectivity of certain targets. In addition, the confirmation that the same gene product is essential in different species greatly increases the confidence in such a target (Freiberg and Brötz-Oesterhelt, 2005).

However, in industry there is a general agreement that well validated, often quite classical targets are not the limiting factor anymore, partly due to novel technologies (Thomson *et al.*, 2004). Rather, the limiting step has become the identification of valuable ligands. Therefore the next generation of genomic based developments should focus, among other things, on functional genomics, enabling the elucidation of the mode of action of specific molecules, as well as toxicity and efficacy prediction (toxicogenomics) of possible ligands.

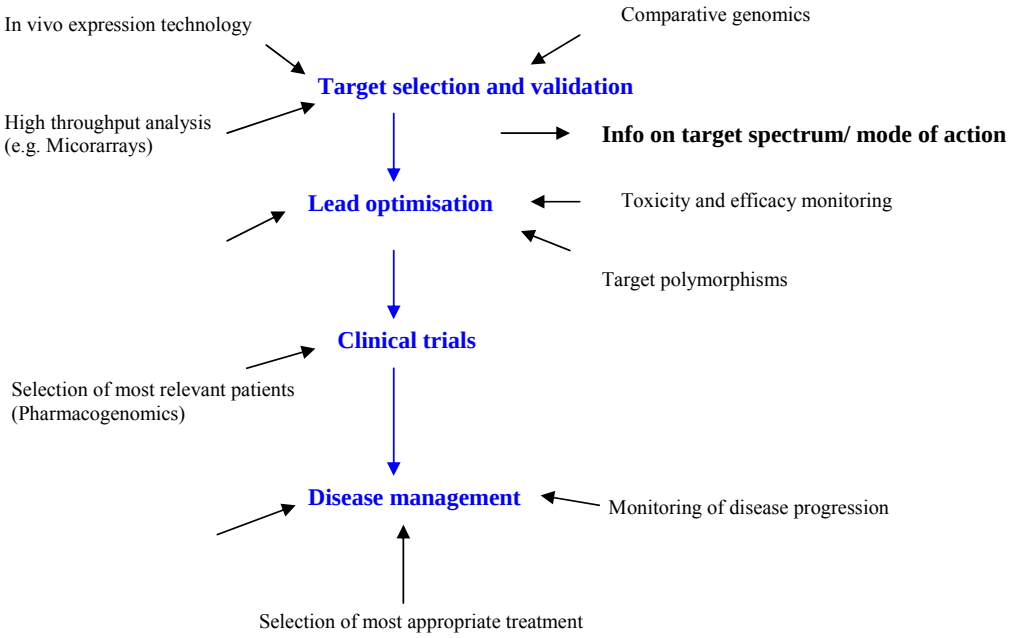

Fig 4: From target selection to disease management – major steps and research methodologies.
Adapted from Impact of genomics on the drug discovery and disease management process. Parkinson, 2002.

Genomics and anti-infectives: main research challenges

a. Target prioritization and validation

Although there is indeed a huge reservoir of pathogenic drug targets, new suitable targets are nevertheless sought for, and therefore genomics based target research should be combined with transcriptional studies, proteomics and fitness profiling. Targets are usually selected based on two criteria, namely conservation among related pathogenic species and lack of a human counterpart. The former, target conservation among pathogens, is obviously required for broad-spectrum drugs. The latter criterion, however, may need some rethinking. For example, the drug fosfomycin is poorly tolerated, although the *murA* gene, coding for a cell wall target, has no clear human homologue. Conversely, azoles that inhibit *ERG11* (encoding lanosterol 14α-demthylase) are considered non-toxic despite a high degree of amino acid sequence conservation between the corresponding human and fungal proteins (Jiang *et al*., 2002). Therefore, genomic studies alone may not be sufficient for accurate prediction of the spectrum or the toxicity of drugs developed against specific targets. Rather, genomic and bioinformatics methods should be extended to include the functional level when evaluating potential targets. To this end, comparative genomics in association with fitness profiling and protein microarrays can greatly enhance our ability to conduct genome-wide functional studies. To date more than eight complete or near complete fungal genome sequences are publicly available (see Useful Links), offering a wealth of comparative information.

Virulence factors should also be considered as potential targets, since targeting pathogenic virulence rather than survival may lead to a reduced selection pressure and, thereby, fewer drug-resistant mutations. Their utility, however, needs to be validated in order to attract the interest of big pharma. A major hurdle in identifying virulence genes is the need to study epidemiologically relevant strains. Indeed, verification of candidate virulence genes must be made in the pathogen of choice and not in a reference strain (very often attenuated or not representative genetically) since a substantial genomic and physiological diversion can exist between the model pathogen and circulating varieties. A possible way to tackle this problem is by signature tagged mutagenesis (STM) (Jiang *et al*., 2002). STM involves the creation of a tagged mutant collection that is pooled and used for infection. Mutant strains that are unable to grow following infection may lead to candidate genes necessary for *in vivo* growth and/or virulence.

Phage genomics (Lui *et al*., 2004) is yet another way to glean valuable information and develop screening tools for novel or unexplored targets. Bacteriophages have developed unique proteins that arrest critical cellular processes in order to commit bacterial host metabolism to phage reproduction. Therefore, phage-mediated bacterial growth inhibitors can form the basis of novel antibiotics. For example, 31 novel polypeptides of *S. aureus* phages were shown to target essential host DNA replication and transcription components. Chemical compounds mimicking these inhibitory polypeptides could potentially function as anti-bacterial drugs (Lui *et al*., 2004).

Finally, high throughput analysis and *in silico* technologies such as virtual screening could be used in order to prioritize identified targets according to different criteria such as "screenability, drugability, and physiological context". Virtual screening could also provide important insights into target-drug compatibility and considerably enhance the chance to find a hit in a given compound library (Knowles *et al.*, 2002).

In this context, the opposite direction for knowledge transfer is also worth considering, i.e. to use molecules that were rejected by the industry (e.g. due to toxicity) for research. Such discarded targets, if made publicly available, can still be of value for the elucidation of biological processes since they are specific and their mode of action is often known.

b. Mode of action

Genomic resources should be better exploited to elucidate the mode of action (MoA) of chemical compounds and natural products as well as for lead optimisation. Target analysis, gene alignments, high-throughput analysis and functional genomics can help to identify active compounds *in vitro*, and provide crucial hints on the MoA of certain hits of yet unknown function (Freiberg *et al.*, 2005). For example, the combination of transcript and fitness profiling led to a better understanding of the MoA of a new class of proteasome inhibitors (Fleming *et al.*, 2002). To obtain the best possible profiles, gene sets whose expression is indicative of the compound's MoA should be identified. Approximately 130 carefully chosen transcripts seem to be sufficient for identifying the MoA of novel substances (Freiberg *et al*., 2006).

Such transcript or proteomic profiles from pathogens exposed to different antimicrobials should be assembled in a common accessible databank, since this information could be used

to define the MoA of other compounds. Finally, databases with a short description of the MoA of known molecules, including natural products, should also be set up.

c. Development of resistance

Drug target selection should take into account resistance mechanisms and frequencies. Notably, resistance-conferring mutations are not limited to target genes, but can also occur in drug transporter genes, bypass genes or regulatory regions. The combination of DNA microarrays and proteomics can help to identify stress-induced proteins that may be non-specifically upregulated by external pressures such as antibiotics, thereby conferring resistance (Hecker and Volker, 2001). If *in vitro* or *in vivo* assays show high spontaneous resistance frequencies, or if alternative pathways to the targeted pathway are known, one can predict problems at later development stages. Therefore, a clear understanding of a compound's MoA is helpful to evaluate the odds of developing resistance and hence the compound's potential to become a long lasting drug. .

To avoid the occurrence of resistance and thus to improve the efficacy of a drug, combination therapy is often recommended. Here too, functional genomics may be of use for predicting the efficiency of such drug cocktails by evaluating the sensitivity of viable knock out (KO) mutants to existing antimicrobials. This requires that KO mutants are catalogued and made available to the community. Important indications for combination therapy could also be derived from synthetic lethal screening, since such screens uncover interactions or redundancies between of two or more proteins.

On another level, drug resistance can be staved off by encouraging the circulation of data concerning the activity of existing and new drugs between clinicians, scientists and pharmaceutical companies. For example, it is important to share information about cross-resistance, or to know if genetic changes in the therapeutic target (genotypic data) can affect drug susceptibility (phenotypic data). This information may allow to predict how pathogens respond to a second line treatment (clinical data).

d. Toxicity and efficiency monitoring

High throughput strategies, such as microarray based technologies, are increasingly used in order to assess toxicity, and recently also efficiency of novel drugs. Biomarker based

strategies for the evaluation of the efficacy of antibacterial compounds have been proposed by Freiberg *et al.* (2006). This study systematically investigated the influence of currently known antibacterial compounds on gene expression by using microarrays. The antibiotic agents used represent the majority of established and commercially available antibiotics (β- lacatams, macrolides, quinolones). The results defined a dataset of some 130 genes that may serve as robust biomarkers for antibiotic efficiency. Such an approach would enable a standardised and automated drug discovery process.

Genomic strategies are also utilized to assess drug toxicity. The "toxico-genomics" approach consists of looking at toxicant-specific patterns of gene expression to identify compounds that will exhibit adverse side effects (Fischer, 2005). Databases of mRNA profiles that reflect treatments with known toxicants, generally agree with what is known from complementary methods such as expression in tissues, and *in vivo* and *in vitro* dosing studies (Fischer, 2005). A similar approach can be implemented at the proteomic level. Such studies can uncover possible toxic effects of a drug candidate at a very early stage and can reveal further details regarding toxic mechanisms (Ulrich and Friend, 2002).

An additional side product of the "-omics" era is pharmacogenomics, a discipline which, on the long term, will lead to a smarter design of therapeutic strategies and clinical trials and to personalised medicine. Within a decade, DNA sequence information will become integral to the practice of medicine. Drug prescription may ultimately be dictated by the patient's genetic makeup (Merrill and Mazza, 2006) as well as by pathogen genomics. This information will also affect the design and execution of clinical trials, culminating in cheaper, faster and more efficient clinical trials, with fewer adverse effects, leading to more rapid approvals.

e. Techniques and models

Transcript profiling has limitations, one being lack of specificity, since typically expression changes in hundreds of genes are simultaneously detected. Efforts should be directed to better correlating between changes in gene expression as a response to a particular condition and the function of such genes. Finally, better methods are needed to distinguish direct and specific responses from the numerous indirect ones. This includes clearly defined bio-assay conditions, and in particular suitable biological models *(in vitro* and *in vivo).* Therefore an inventory of models should be assembled according to the biological disease studied, taking into account: i) the essential interactions between host and pathogen, and ii) the essential

molecular interactions enabling survival of the pathogen within the host. Existing models should be adapted (e.g. humanised mice) in order to minimise the differences between reality and model. Biomarkers could be of great value in this context, by comparing disease-related human markers to the markers observed in the animal or cellular model. Identification of significant markers requires, of course, detailed knowledge of the disease at the human level and a close collaboration with clinicians.

In the context of drug discovery, a good model should accommodate the molecules implicated in a particular MoA, and supply information regarding their role and the consequence of disrupting their respective molecular pathways. This would consequently give an added value to the drug candidate under examination and will facilitate the attraction of appropriate partners for the developmental process. In the context of the Innovation process, the following issues should be clearly and accurately formalized in order to select the appropriate model: i) what are the fundamental questions to be answered according to the studied pathogen, and ii) which questions need to be addressed at the pre-clinical stage according to industrial partners (e.g. for target validation). For example, whole-cell-based reporter assays have limitations, particularly as a result of the limited concentration window in which compounds might be detected as inducing agents (Freiberg and Brötz-Oesterhelt, 2005).

As a complementary approach to model optimisation, novel *in silico* methods will undoubtedly be developed to reconstruct regulatory networks, signalling cascades and metabolic pathways, with an emphasis on comparative genomics and microarray-based data. The current bottleneck however, lays in the biological integration and interpretation of the data, the lack of tools to reconstruct complete disease pathways, and lack of predictive models to simulate the dynamics of biological systems during disease and treatment conditions (Fischer, 2005).

On a more mundane level, computer based systems play an increasingly important role in storing, structuring and analysing research data, and it is therefore extremely important to instate a uniform, standardised coding system. The system should allow to share, distribute and compare findings, especially since future research seems to evolve more and more towards multidisciplinarity. Since far reaching decisions are increasingly based on automatically generated data, specialised quality control systems are being developed that can spot erroneous data which might obscure important biological effects. It should be

emphasized, though, that biological interpretation of the data remains crucial for identifying and validating drug targets. Therefore qualified people remain an essential link in the drug identification process.

Despite their promise, it looks like the "-omics" didn't usher the expected "golden age" for new antibiotic research. They did, however, considerably improve drug discovery efficiency, making bacteriology one of the rare disciplines where pre-clinical data can predict, to a certain extent, efficacy in clinical development. After lead compounds are chosen, functional genomics is extremely helpful also for rapid assessment of their potential early in the clinical development process. This means that there are fewer candidate antibacterial compounds entering clinical development than in other disciplines. Unfortunately, this is often perceived as a weakness (Thomson *et al*., 2004), but in fact, even more efforts should be put into the establishment of "tailored" compound collections that have a potentially increased rate of success. This could be done, for example, by identifying features enabling compounds to cross microbial membranes, either by comparing effective and ineffective compounds, or by using chemoinformatics. Due to the high diversity of bacterial strains and pathologies, antibacterials should be available in a wide range of formulations (oral, parenteral, etc.), adding a level of complexity, which deserves additional attention (Thomson *et al.,* 2004).

Genomics in anti-infective research and tools – summary

General needs

* A uniform, standardised coding system of genes, proteins, mRNA-sequences
* Integration of quality control systems in databases to spot erroneous data

Target validation and inhibitor compatibility

* Need for scalable database systems and computational tools for target discovery
* Target prioritisation should be extended to the functional level by:
  - comparative genomics, in association with fitness profiling and protein microarrays (HTS)
  - phage genomics
  - *in silico* technologies such as virtual screening methods for target-inhibitor compatibility
* Virulence factors should be considered as possible targets (use of a combination of genetic and molecular tools for validation).

* Better integration of epidemiology and population genomics: integration of molecular strain data with epidemiological and clinical data

Mode of Action (MoA)

* Determination of the MoA should be extended to the functional level by:
  - combination of transcript profiling and fitness studies
  - creating an inventory of transcripts regulated by known molecules to help determine the MoA of novel hits
  - organising a common databank containing the transcript or proteomic profiles of pathogens exposed to different antimicrobials used in whole cell screens.
* Creating a database of MoA of known molecules.

Development of resistance

* More exhaustive determination of direct and indirect resistance mechanisms by using protein microarrays, synthetic lethal screening and other methods
* Use of functional genomics for the evaluation of combination therapies
* Catalogue available knock out mutants
* Organise a common databank listing cross-activity and resistance of existing drugs against circulating species and strains. This would give important insights on the consequences of genetic changes at the phenotypic and clinical level.

Toxicity and efficiency monitoring

* Better integration and further development of *in silico* methods at the proof of concept level e.g. toxicogenomics or efficiency monitoring by analysing high throughput screening results with the appropriate bioinformatics tools
* Use of genomics for the identification of biomarkers for efficiency monitoring.
* Establish a database of identified biomarkers available from academia and industry
* Organise a database with protein profiles or mRNA profiles associated with toxic effects of known molecules

Techniques and models

* Better *in vivo* models: delineate guidelines for good animal models
* Make an inventory:

- according to the disease studied
- according to the scientific questions and biological context

- according to the pre-clinical development needs

* Establishing sets of biomarkers for monitoring disease development and progression in humans and animal models.
* Develop tools for differentiating specific responses from non-specific ones when using high throughput systems.
* Need for i*n silico* methods to reconstruct regulatory networks, signalling cascades and metabolic pathways to create predictive models, while using a uniform, standardised coding system.
* Lobby for redirecting compounds rejected by the industry back to the academia, to be used in basic research
* Create a database of compounds which are able to cross the microbial membrane, thus enabling the identification common features and optimising antibacterial and antifungal compound collections
* Need for more research on right delivery systems for the right pathogen (oral, parenteral, etc.)

Political and legal needs and solutions

To support anti-infective R&D, important political input is needed. Indeed, it is up to federal agencies to implement programs for educating clinicians and the public on the appropriate use of antimicrobial agents. Such programs should find the balance between the need for reduction in antibiotic usage and the need for development of new agents.

To modernise and revitalise the drug development process, the FDA (US) has drafted a list of priority projects for developing new tools that could significantly improve assessment of experimental drugs (part of the Critical Path Initiative). Six broad topics are covered: development of biomarkers, clinical trial designs, bioinformatics, manufacturing, public health needs and paediatrics (www.c-path.org). Success will require extensive collaborations between the government, academia and the industry. Some companies are already interested in joining forces (e.g. in sharing laboratory methods to predict the safety of new treatments

before they are tested in humans), but funding remains a challenge since the FDA limits financial input of the industry, thereby leaving a financial gap.

A similar initiative should be introduced in Europe. Due to the general financial anaemia on the European Stock Exchanges and the paucity of European investors in technology stocks, solutions for revitalising anti-infective R&D could include the following points**:**

Political and legal needs at the European level - summary

a) More collaborative programs for antibacterial drug R&D involving European authorities, academia and industry, in order to balance between applied and basic research.

b) Shorten the time between deposit of proposals and actual financing (6 months) of SMEs, since ~1 year is too long for an R&D initiative.

c) Explore the possibility of governmental or EU contracts with industry to develop antibacterials for specific needs

d) Streamlining and unifying patenting procedures, with the goal of creating an EC patenting procedure that will be valid simultaneously in all the community countries.

e) Initiating legislation providing economic incentives for the anti-infective industry, such as tax incentives.

f) Improved communication between the regulators (EC), industry, competence centres and Member States policy makers (ministries) in order to amalgamate research in Europe.

g) Enable access to anonymous clinical files, strictly for scientific use.

h) Discuss possible solutions to problems in managing clinical trials and recruiting enough volunteers.

i) Increase efforts to streamline the drug approval process without compromising safety and efficacy standards.

j) Synchronize European tariffs, thus avoiding lengthy negotiations with regulators and health care agencies of each Member State and speeding up the marketing process.

The European Commission already implemented or plans to implement new guidelines to overcome existing problems in the European pharmaceutical industry and to asses its competitiveness. Some of these policy changes include:

- Faster market access for products offering significant therapeutic benefits, through an accelerated procedure and the possibility of conditional approval for breakthrough treatments.
- Streamlining the regulatory process and directing EMEA (European Agency for the Evaluation of Medicinal Products) towards providing scientific guidance and support to industry (SME's) (http://www.europabio.org/articles/PR_SMEs%20and%20EMEA_FINAL.doc).
- Clarifying market exclusivity status by defining a ten year data exclusivity period (with an additional year granted for innovative research on already marketed products). Generic applicants are allowed to prepare for marketing before data exclusivity expires.
- Assembling thematic programs proposing international calls (Era-Nets), in order to coordinate the different research centres and national policy makers.
- A clinical trial directive has been instated, establishing principles of Good Clinical Practice (GCP), aimed at improving coordination of trials across Europe. Also, a European clinical trials database has been established to enhance communication between regulatory agencies.
- To promote a greater visibility of European SME's, EuropaBio created Channel Bf (www.channelbf.com), a new online channel offering operators and companies different formats to present themselves online.
- The European Commission launched a novel initiative, the Innovative Medicines Initiative (IMI) (http://ec.europa.eu/research/fp6/index_en.cfm?p=1_innomed). The overall objective of IMI is to remove bottlenecks in the development of new medicines, positioning the European biopharmaceutical industry as a world leader. At the same time, by pooling resources from all stakeholders (industry, academia, SMEs, regulatory authorities, healthcare providers, patient organisations), this initiative is expected to provide faster access to better medicines for European citizens. This will consist of a call for proposals regarding efficiency and toxicity predictions in the field of infectious diseases.

Intellectual property, systems biology and multidisciplinary approaches

An important question is, how the field of IP rights will evolve in a research environment emphasising complex, multidisciplinary research rather than focusing on individual molecules or mechanisms. It is expected that knowledge will be pooled from the public domain or from

patent holders willing to trade at a reasonable cost, which makes an affordable pricing of the final product a possible future challenge.

An additional issue to consider is the possible restriction of the emerging field of systems biology due to patenting of large research areas. This may have significant impact on the technological opportunities available for follow-on developers (Allarakhia & Wensley, 2005). Dominance of patents filed earlier in time, at the most fundamental levels of the biological information hierarchy, can therefore significantly hinder the incentive to conduct research in the higher levels of the hierarchy (Heller & Eisenberg, 1998)

Intellectual property professionals are unsure whether the value of biological pathway is equivalent to discovering and developing a drug that acts on this pathway to effectively treat a disease. In other words, is the discovery of a biological pathway sufficient in itself to merit the granting of a broad patent that claims any treatment acting on that pathway? An example is the case of the nuclear factor κB (NF-κB) cell signalling pathway. A patent application was filed by different institutes, but an exclusive license was granted to a particular pharmaceutical company for all patents related to methods of treating human disease through modulation this particular pathway. Such patents could be used by the owning institutes against other researchers, blocking off competing research in the attempt to commercialise their discoveries (Allarakhia & Wensley, 2005).

To avoid appropriation of such important biological pathways, it is of uttermost importance that a political and legal framework should be setup to ensure that research and patenting remain accessible to all research groups. Whether or not systems as a whole should be patentable remains a major concern.

III.c. Disease monitoring and prevention.

Impact of microbial genomics on control of infectious diseases: further needs

A diagnostic test for infectious diseases should indicate, directly or indirectly, the presence or absence of a pathogen. Monitoring tools are meant to give information on disease progression, such as stage of the disease, resistance monitoring, effectiveness of treatment, etc. The importance of diagnostics is increasingly recognized, leading to a greater demand for faster diagnosis and more markers for disease monitoring. Outbreaks of avian flu, or of the SARS-

virus, have highlighted the necessity for such tools. Consequently, the field should be undergoing an important growth.

Advances in biomedicine are forging a link between genomics (genetics, gene expression, etc.) and disease diagnosis, monitoring and treatment in a way that was not possible before. Infectious agents will be detected faster, with greater accuracy and at earlier stages and disease progression will be easier to follow. Moreover, diagnosis directly from clinical specimens will be possible, avoiding possible artifacts by culture conditions. Consequently, the treatment of infected patients should be optimized.

Despite a clear need for such tools, the fact that most of the technology is available and the relatively low development costs of diagnostic and monitoring kits, it is sometimes difficult for new diagnostic companies to attract investors and prosper. Therefore it is necessary to increase awareness of such tools, coordinate research, and assemble scientific databases. Political backup from policy makers lobbying for involvement of investors and industry is also necessary. In this section, some of the remaining gaps in microbiological identification and monitoring will be highlighted, ongoing actions will be described and possible solutions suggested.

III.c.1. Pathogen diagnosis and monitoring tools

The ideal identification of a causative agent is probably a combination of several methods (nucleic acid amplification, biochemistry, immunology and culture methods), specifically tailored to a particular issue or need (degree of identification, evidencing of resistance, speed, cost, etc.).

The level of detail of pathogen identification (ranging from strain to genus) may vary. For epidemiological tracing (e.g. for identification of the source of contamination), the exact strain is important, whereas for treatment it is sometimes enough to identify the genus. Identification of resistant pathogens can be done empirically, by verifying persistence after treatment, or by confirming the presence of resistance related mutations or markers.

The modern diagnostic test is expected to be fast (results within a few hours at most), sensitive, specific, easy to use (4-5 steps), cheap and scalable.

a. Specific needs

Direct identification of a pathogen is done by detecting a particular target (gene sequence or protein), preferably using the most specific, sensitive and simple assay. However, there are a number of pathogens and conditions for which rapid and specific diagnostic tests are still lacking: *Asperguillosis* in immuno-depressed patients (Kontoyiannis, ECCMID, Nice), Mycobacterium Tuberculosis (pulmonary and extra-pulmonary), meningitis-encephalitis differentiation, early sepsis detection, agents causing respiratory tract infections, infant diarrheal infections (virus vs. bacteria) and agents causing endocarditis.

Additionally, rapid screening tests for various multi drug resistant organisms are painfully lacking. These include penicillin-resistant *pneumococci* (Leclercq, ECCMID, Nice), methicillin resistant *S. aureus* (MRSA), vancomycin resistant enterococci (VRE) and extended spectrum beta-lactamase (ESBL) producing strains (Robin *et al*., 2006). Even when tests are available, false negatives constitute an ongoing problem (Leclercq, ECCMID, Nice).

In general, there is an urgent need for tools identifying resistant strains (see also section III.c.1f - resistance monitoring) and tests detecting latent infections. The latter are mainly important to identify people at risk for developing an active infection and the identification of potential reservoirs (e.g. for Mycobacterium infections).

b. Epidemiological needs

For epidemiological and population genomics purposes, more universal targets (present in all studied pathogens) are used. The most commonly used universal gene targets are the 16S rRNA gene, the 16S-23S rRNA intergenic spacer regions and the 23S rRNA locus. These are highly conserved and found in all bacteria. The only commercially available identification kit for a universal gene is based on the 16S rRNA locus (MicroSeq 500 16S ribosomal DNA bacterial sequencing kit from Applied BioSystems, Millar and More, 2004). Possible alternatives to the ribosomal sequences include the rpoB gene encoding the β subunit of the DNA-dependent RNA polymerase (Ait Tayeb *et al.* 2005) and heat shock proteins (HSP).

A different way of strain-typing is by Multilocus Sequence Typing (MLST) using sequences of internal fragments of seven house keeping genes, which can be retrieved from the central MLST bank (http://www.mlst.net/misc/further.asp). Allelic profiles of isolates can be

obtained from clinical material by PCR amplification directly from the CSF or blood (Taha *et al*., 2005).

More recently, Single Nucleotide Polymorphisms (SNPs) have been used for genome-wide analysis of different strains (recombination, chromosomal dynamics, genome rearrangement). This method is suitable for genotyping and population genetics studies, and is extensively used for human genetics and the detection of predisposition to disease in humans. Only two companies (Affymetrix and Illumina) provide high-throughput genome wide genotyping tools applicable for the research laboratory, (more than 100,000 SNPs in a single experiment), but only well-funded laboratories stand to benefit from such kits. Although infectious disease genotyping requires much less SNPs (50-100 SNPs), only one commercial kit is available (SNPlex ™ from Applied Biosystems, 48 SNPs/sample). SNP typing is one of the most discriminative tools available at the moment and it could be a powerful tool also for disease monitoring (see III.c.1e - *disease monitoring tools*). Yet it should be noted that this method is a relatively time-consuming, multi-step procedure. Therefore, novel techniques and identification methods are still needed in order to detect and monitor the spread of newly emerging diseases.

Additionally, there is a need for centralization and standardization of sequence nomenclature and of the different public sequence databanks. Indeed, it is often quite trying to identify all known sequences of a particular gene or region. Although some commercial solutions exist (NaviBio ™ and GetDB ™ from Genomining), open-access solutions should be sought.

Finally, sequencing of whole genomes is constantly becoming faster and cheaper. Consequently, special attention should devoted to programs able to compare whole genomes, and monitor genome and virulence factor evolution, in order to study the epidemiology of specific infectious agents (Beres *et al.*, 2006).

c. Technological needs

Genome based detection methods require proper sampling of genetic material, and often elaborate sample preparation. Therefore, simplification of the preparation procedure is warranted. Additionally, isolation and stability of RNA remain problematic, especially from faeces, the starting material for detecting pathogens such as *Salmonella typhi* and *Shigella* spp. The ideal diagnostic test should also take into account the pathology and progression of

the disease as well as pathogen-host interactions, in order to allow efficient sampling with minimal invasiveness and maximal sensitivity.

Novel sequencing techniques should help render disease detection and monitoring faster and more accessible. One such example is pyrosequencing (Ahmadian, *et al.*, 2006), a promising and very fast technique (1 hour), albeit limited, for the time being, to short sequences (60-100 bp). A commercial pyrosequencing kit is already available (PyroMark ID, http://www.biotagebio.com/DynPage.aspx?id=30490), and can be used for species identification (bacteria and fungi) and resistance detection.

Finally, apart from the technological limitations, the high price of an assay remains in many cases a major bottleneck. With the advances of technology, the increase in large scale production and the expiry of certain patents, costs will eventually drop.

Some examples of open access databases are:

- PulseNet (http://www.cdc.gov/pulsenet/): bacterial food-borne disease surveillance in the US
- ESF Network for Exchange of Microbial Typing Information (ENEMTI) (http://lists.nottingham.ac.uk/mailman/listinfo/enemti)
- European Society for Clinical Microbiology and Infectious Disease (ESCMID) specific Study Group on Epidemiological Markers (ESGEM)

Formatiert: Nummerierung und Aufzählungszeichen

d. Standardization of diagnostic tools

To advance diagnostic tools from the research lab to the market, tests need to be adapted and validated. Molecular and serological diagnosis and detection methods require the use of standardized materials and approval by international quality control programs. Currently, however, diagnostic tools are subject to a somewhat lax set of controls and regulations, especially when compared to the very strict legislation and tests concerning the development and use of new drugs.

Due to this inadequate standardization and regulation, there has been a proliferation of in-house diagnostic methods, only some with significant value to the scientific community. As a result, diagnostic surveys must often include a preliminary analysis of published work, comparing PCR primers, antibodies, and other materials used. As a result, more and more scientists are comparing and coordinating PCR assays (Taha *et al.*, 2005; De Baere *et al.*, 2005), an important initiative that should be encouraged. There is also little uniformity in the genetic makeup of organisms, and little attention paid to possible cross-reactions. Rarely do commercially available tests address the issue of cross-reactivity between strains. Even if there is no cross-reactivity with one particular reference strain (that might not even accurately represent of the whole field), cross-reactivity with other strains of the same species cannot systematically be ruled out. Additionally, diagnostic tests often fail to consider strains endemic to developing countries, as was the case for certain African HIV strains in the past. Detection techniques should cover genetic diversity as broadly as possible, especially in light of the flourishing of genomic approaches in the study of population genetics, taxonomy and epidemiology.

Since an accurate picture of the natural diversity of strains and species is crucial for designing a successful diagnostic test, close collaboration of taxonomists, population geneticists and epidemiologists is recommended. One outcome of such a collaboration may be a fixed panel of organisms to be tested for each pathology. This panel should be freely available as a sero-library or cryo-bank to any group willing to evaluate the value of the developed test. In parallel, well documented samples should be available for clinical and statistical validation. This would make it much easier to compare the efficiency of various diagnostic tests, and would lend further credence to existing ones who already show good results.

Although the European Community requested a Reference Measurement System (RMS) to be set up for each and every quantity measured or determined in laboratory medicine, many gaps remain in the standardization of lab monitoring tools (Lequin, 2003). Other initiatives towards standardization are being established in international congresses (ECCMID, Nice, Educational workshop on Quality control and standardization in molecular diagnostics, by the ESCMID study group on Molecular Diagnostics) and by organizations outside Europe, such as the Clinical and Laboratory Standards Institute (USA) (e.g. Molecular Diagnostic Methods for Infectious Diseases: approved Guidelines, http://www.nccls.org/Content/NavigationMenu/NewsEvents/DownloadMarketingBrochures/

ASTBrochure106.pdf) However, the European Community should still initiate and coordinate a universal action towards monitoring of the entire diagnostic procedure, thus guaranteeing the consistency and accuracy of the results generated.

e. Disease monitoring tools

One of the exciting outcomes of genomics and the related novel high throughput molecular techniques, is the possible identification of microbial markers. Markers can help monitor disease progression and the efficacy of medical interventions. These markers can be chemical substances, proteins or specific transcripts. Monitoring disease progression includes the identification of specific microbial and host factors during the various stages of human clinical disease and should enable a temporally adapted treatment.

Markers can be particularly useful in progressive diseases, involving phases such as colonization, invasion, disease development and inflammation and possibly septic shock. Such difficult to treat systemic infections can be caused by various pathogens such as streptococci, *E.coli, Klebsiella, Enterobacter, S. aureus, Listeria, Candida albicans* and more. Sepsis is a spectrum of clinical conditions caused by an immune response to infection that is characterized by inflammation and coagulation which can lead to organ failure. Therefore early detection of a septic response can save lives. But since sepsis describes the magnitude of the host response to the infection and not the infection per se, reliable detection is challenging. One objective, then, is to find the correct balance between a simple and fast procedure (using a minimum of markers) and a reliable one, especially when using indirect markers.

Another disease with gradual progression is tuberculosis, caused by *Mycobacterium tuberculosis,* an intracellular pathogen capable of surviving and persisting within host mononuclear cells. A subclinical latent or chronic infection may persist even in healthy hosts (Gomez & Mc Kinney, 2004) but neutralization of Interferon gamma (IFNγ) or Tumor Necrosis Factor (TNF), inhibition of iNOS (IL induced nitric oxide synthetase) or T-cell depletion leads to reactivation of latent infection (Botha, 2003). Specific markers for disease progression are therefore needed for early detection and appropriate treatment.

SNP analysis could identify such markers, as exemplified for tuberculous meningitis, an extrapulmonary form of Mycobacterium tuberculosis (Hawn *et al.*, 2006). Specific polymorphisms in an adaptor protein that mediates signals from Toll-like receptors activated

by mycobacteria, have been shown to be associated with increased susceptibility to tuberculosis and particularly to meningeal tuberculosis (Hawn *et al.,* 2006).

Another approach for marker identification is proteomics. Proteomic studies were used to map Candida albicans immunogenic proteins specifically recognized by antibodies produced during systemic Candida infection. This approach has led to the characterization of C. albicans antigens that are associated with a differentiation of the human immune response. In addition, antibodies against other Candida antigens were found to correlate with recovery from systemic candidiasis. Such markers can serve to predict the preferred treatment and outcome of the disease (Pitarch *et al*., 2004). Such findings have to be confirmed by large scale studies, and technological solutions are needed for identifying these markers in a fast, robust and simple way. Be that as it may, such studies demonstrate that thorough knowledge of the host-pathogen interaction is necessary for identifying such valuable monitoring tools.

Biomarkers can also be used as indicators of efficacy and safety during treatment interventions in pilot studies (see also III.b. anti-infective research and tools). This could reduce the number of efficacy studies required for each additional indication, and consequently the costs, while maintaining safe and effective drug dose regimens. Biomarkers are already increasingly used for making informed development decisions in the industry (e.g. for early clinical proof of concept).

To identify the best possible markers it is very important to have access to clinical material and files and a good communication between scientists, who know the biology of the pathogen, and clinicians, who have a clear picture of disease progression. Epidemiological and clinical data are critical, as well as informative animal models who mimic the infectious process.

Due to the fast pace of molecular tool development, science is far ahead of legislation. Legal validation of biological markers should be speeded up by legislators and policy makers. For example, a clear definition of acceptable surrogate markers as end points for clinical trials of bacterial infections is lacking. Once the needs are clearly defined, studies could focus on identifying relevant markers and assessing their levels at different stages of the infection.

f. Resistance monitoring

The rise and spread of drug resistance is an ever increasing problem, since second-line medication is not always available, and resistance increases the cost and complexity of treatment. Early-stage detection of resistant pathogens can save time, energy and financial resources, and yet in most cases rapid, cheap tests to detect the presence of a resistant agent are not available. The ideal test should identify the presence of viable, resistant pathogens directly in a patient's sample. With presently available techniques, this means demonstrating specific antigens or nucleic acid sequences. Detection of nucleic acids should preferably focus on RNA, since it is not yet clear how long does DNA persist in the blood after treatment. Genome analysis and comparative genomics techniques can help to identify direct or indirect drug resistance markers. Commonly used markers are mutations such as those found in QRD Regions (quinolone resistance-determining regions) and in gyrA, gyrB, parC, parE genes (Ip *et al*., 2006). Such markers are very useful for the identification and monitoring of resistance among circulating strains. Other genome-wide techniques such as SNP analysis could supply insights into the resistance mechanisms by detecting sequence heterogeneity between resistant and susceptible organisms. This technique could possibly also provide some indirectly linked resistant markers (Coste *et al.*, 2006).

Novel *in silico* genome methods can reveal molecular mechanisms of resistance to several classes of antibiotics within drug targets. For example, it is possible to predict resistance of the *Ricketsia* bacteria to several classes of antibiotics by genome comparisons analyzing mutation profiles in key molecules (e.g. rpoB genes, related to rifampicin resistance) or by looking at the presence/absence of drug target molecules and sequence homologies (Rolain and Raoult, 2005). Such methods can be very useful particularly for intracellular bacteria for which transformation assays are usually not suitable,.

A novel initiative, GRACE (Genomics to combat Resistance against Antibiotics in Community acquired low respiratory tract infections in Europe), was recently launched with support from the European Sixth Framework Program. Lower tract infections (LRTI) are a leading cause of antibiotic use and contribute dramatically to the rising prevalence of resistance among major human pathogens. The overall objective of GRACE is to combat antimicrobial resistance through integrating centres of research excellence and exploiting genomics in the investigation of community-acquired LRTI. Microbial and human genomics will be integrated with clinical observational and intervention studies, to specifically change

practices used for managing community-acquired LRTI. To this end, GRACE will encourage development of novel, rapid genome based diagnostic tests for the detection of pathogens implicated in community-acquired LRTI.

However, before sophisticated techniques can be used, some important standardizations should be implemented. First, open access, uniform databases, with standardized nomenclature should be established. An example is the Stanford HIV drug resistance database (http://hivdb.stanford.edu/). Second, drug-susceptibility tests should be standardized and calibrated such that results are reproducible in different reference centers, which currently is not always the case (Kim, 2005).

The genomic era initiated large-scale identification and validation of biomarkers and drug targets. The vast amount of data generated should be deposited in open access databases, were profiles, sequences etc. could be accessed and compared. The acquired information (genomic, proteomic) will be used for the development of fast, serological or molecular tests, with the possibility of quantification. In the near future, new technologies such as nanotechnologies, multiplex PCR techniques, optimized array techniques (optical fiber arrays, Walt, 2006), and spectrometry, should be incorporated.

Identification of pathogens - summary

Specific clinical needs

* need for better and faster diagnostics for:
  - penicillin-resistant *pneumococci*
  - more specific MRSA, VRE and ESBL detection tests
  - broad spectrum sensitive assays for *Asperguillosis* in immuno-depressed patients
  - Mycobacterium tuberculosis infections (active, latent and extrapulmonary infections)
  - to differentiate meningitis/encephalitis
  - early sepsis diagnosis
  - respiratory tract infections
  - infant diarrhea infections (viral vs. bacterial)
  - identification of endocarditis agents
  - detecting latent infections
  - detecting nonculturable pathogens

Epidemiological needs

* simplification of the procedure.
* further development of genome comparison programs.
* affordable, infectious diseases adapted, user-friendly SNP packages.
* standardization of sequence nomenclature.

Technical needs

* better methods for extracting DNA/RNA from clinical samples, especially form faeces.
* better sequencing methods:
  - faster
  - cheaper
  - output more user friendly
* cheaper DNA/RNA based methods

Need for standardization

* determination of a regularly updated, open access, fixed diagnostic panel of organisms, that are tested for sera specificity (cross-reactivity), sensitivity and epidemiological representativeness.
* well documented human samples should be available for clinical validation.
* need for networks between the different reference centers.
* statistically significant panels should be established for validation.

Disease monitoring tools

* large scale studies to identify microbial factors which are expressed during the various stages of infection and illness in order to identify surrogate markers.
* development of genomic and proteomic based technologies for further detection of such markers and for large scale studies elucidating the host response and the corresponding pathology.
* a legal framework within which biomarkers can be used: when can a marker be considered as an endpoint of a clinical trial, how many and which markers indicate the need to switch treatment, etc.

* clearly defined criteria for marker validation models, and for incorporating candidate markers into a monitoring test.

Resistance monitoring

* better tools are needed to detect resistance and persistence.
* study on pathogen DNA/RNA persistence in blood after treatment.
* development of *in silico* methods to reveal and predict molecular mechanisms underlying resistance.
* standardization and calibration of drug-susceptibility testing procedures (determine a consensus (European) cutoff level for resistance).
* standardization of databases and nomenclature.

General needs

* regulations for test approval should take into account clinical utility of diagnostics
* centralized, accessible databanks (sequences, clinical data, activity of drugs against circulating strains, cross-reactivity).
* centralized, publicly available list of diagnostics, sorted according to lab, purpose, results and price.
* a framework for storing samples for retrospective analysis and other, as yet undefined uses, while paying attention to ethical and legal issues.
* a regulatory framework for co-developing drugs with diagnostics (define the role of academics, industry and consortia).
* more interaction between biologists and clinicians
* identify and prioritize key organisms, diagnostics and monitoring tools.

NEED FOR A GLOBAL FRAMEWORK CONNECTING SCIENTISTS, CLINICIANS, AND EPIDEMIOLOGISTS FOR THE STUDY AND TREATMENT OF INFECTIOUS AND EMERGING DISEASES

III.c.2 Vaccines

Effective vaccines stimulate the body to create antibodies or cellular immune responses to kill or neutralize the disease-causing organism without any side effects and provide immunity for a long period of time. Vaccines are estimated to save three million lives every year (Global Industry Analysts, 2005) and are no doubt a major global health tool.

The vaccine market holds considerable financial potential, especially the therapeutic vaccine sector. Pediatric vaccines, vaccines for emerging diseases and travel vaccines are particularly expected to expand in the near future (Global Industry Analysts, 2005). There are 200 companies worldwide developing more than 600 vaccine products, but the marker is clearly dominated by a few "leaders": Sanofi-Aventis (27.4%), GSK (24.4%) and Wyeth (20.5%) (Global Industry Analysts, 2005). These companies maintain their lead in this competitive market due to long-term expertise, regulatory experience, by establishing vaccine subsidiaries in new geographic regions and by the acquisition of small local partners.

Vaccine development in infectious diseases entered a new era when the first genomes of human pathogens were sequenced. Prophylactic and therapeutic vaccines have rapidly evolved based on the improved genomic and proteomic knowledge. There seems to be a shift from whole cell attenuated vaccines towards antigen specific vaccines with the use of novel technologies for vector design, DNA vaccines and combination vaccines.

The availability of the sequence of all proteins encoded by the pathogen made it possible to design vaccines *in silico*, without the need to grow the disease-causing microorganisms, in what is now called "reverse vaccinology". This approach is particularly useful for diseases caused by unculturable or by highly mutable microorganisms. It has become possible to develop vaccines based on epitopes derived from the whole genome. In theory it should be possible to design a totally synthetic vaccine containing strings of the best epitopes encoded by the targeted microorganism. Poorly immunogenic antigens and antigens who potentially cross-react with human proteins could be excluded. Genome based approaches have been described for vaccines developed against *Neisseria meningitis*, *S. pneumoniae*, streptococcus and *Chlamydia* (Fraser and Rappuoli, 2005). The fast response to the SARS outbreak exemplifies the ability of reverse vaccinology to respond rapidly to emerging infections (Burkreyev *et al.*, 2004). Results of ongoing clinical trials will soon supply information regarding the practical value of reverse vaccinology (Fraser and Rappuoli, 2005).

a. Public health needs

One major public health unmet need is an exhaustive inventory of pathogens susceptible to vaccination. Some high-priority vaccination objectives include the worldwide problem of *Mycobaterium Tuberculosis*, an effective vaccine against *Neisseria meningitis* subtype B (50% of the meningococcus cases) and vaccination as a weapon against resistant bacteria such as MRSA. A vaccine that specifically targets antibiotic resistant bacteria could prevent their spread and thus reduce the antibiotic use and reduce future resistance (Tickell, 2005). However, a complete market analysis on the subject is needed. Additionally more combination vaccines should be considered since they reduce the number of interventions and thus also the costs (Datamonitor, 2005).

b. Research and Development needs

The majority of vaccine research and development has shifted towards peptide vaccines. This type of vaccine is preferred due to its relative ease of construction and production, chemical stability and lack of oncogenic or infectious potential. Some improvements however, are necessary due to i) the poor immunogenecity of peptide vaccines ii) the need for better adjuvant and carrier systems, and iii) the need for reliable and simple assays to measure T-cell response.

The success of a vaccine developers relies on the right combination of antigen, adjuvant and delivery method (Datamonitor, 2005). Further efforts should be put into the discovery of the right antigens and emphasis should be laid on the host-pathogen interaction and on understanding of the role of genes and proteins in complex immune responses and chronic disease progression.

Genomics contribute to the detection of new antigens by allowing to compare two or more genomes. For example, comparison of the Bacille Calmette Guerin (BCG) and *M. tuberculosis* genomes uncovered many genetic differences that may be responsible for the incomplete protection of the BCG vaccine against tuberculosis (Sherman *et al.,* 2004). Genome comparisons can also be used to select B and T cell epitopes, based on computational immunology. Algorithms for T-cell and B-cell epitope mapping are used to scan genomes for major histocompatibility ligands and predict their affinity to allow the design of epitope-based

vaccines containing only the selected subsequences (De Groot, 2006). Complementary population genetics studies should allow the study of the epitope stability in the pathogen population of interest.

Resulting data could also be used to develop new diagnostic tests (such as the ELISpot assay kit, which differentiates tuberculosis infection from BCG vaccination, De Groot, 2006). Comparison of T-cell epitopes of different pathogens might unveil significant differences between pathogens, regarding the number of epitopes that are presented to the immune system. Such studies could clarify host-pathogen interactions. For example, Helicobacter pylori evasion of the host immune system can be explained by similarities between the bacterial and host epitopes (De Groot, 2006).

Immunomics is a new field that addresses the interface between host and pathogen proteome, bridging informatics, genomics, proteomics immunology and clinical medicine. Antigen discovery platforms will help propel this field, and should build upon a combination of technologies such as bio-informatics, spectrometry, *in silico* methods, etc.

Due to the poor immunogenicity of peptide vaccines, more effective antigen delivery technologies, such as vectors and adjuvants, should be developed. The low immunogenicity stems from lack of innate immunity signals activating the adaptive immune response. Innate immunity receptor agonists are therefore a natural choice as adjuvants. Toll-like receptors (TLR), for example, are expressed on innate immunity cells and play an important role in its activation (e.g. TLR mediates maturation of dendritic cells into potent antigen-presenting cells). Indeed, most TLR agonists are good candidates to enhance pro-inflammatory, antibacterial TH1 type responses, because they activate type I Interferon and the NF-κB pathway (Romagne, 2007). Some of these compounds are already present as adjuvants in commercial vaccines such as the hepatitis B vaccine, Fendrix ®, or in clinical trials of prophylactic (malaria and tuberculosis, Persing, 2002) and therapeutic vaccines. However, further research is needed to elucidate the structure-function relationship between the different agonists and their receptors. Indeed, a safe and efficient use of the different agonists requires a better understanding of their biological activities as well as their exact tissue distribution, metabolism and cell uptake (Romagne, 2007).

c. Legal issues

Licensing of antigens will become increasingly important and challenging as well-known antigens are progressively exhausted. Multi-antigen vaccines are expected to become more common, requiring the integration of various technologies and potentially even combination of antigen portfolios from different companies. Consequently, a legal framework enabling such joint ventures should be set up to ensure affordable end products.

Vaccine development - summary

Specific Public health needs

* vaccines that specifically target antibiotic resistant bacteria
* *Neisseria meningitidis* vaccine including the B-type
* TB vaccine
* more combination vaccines

Research and development needs

* an ongoing search for optimal antigens:
  - development of antigen discovery platforms integrating technologies such as bioinformatics, 3D analysis and *in silico* techniques
  - developing effective antigen delivery technologies
  - choosing efficient adjuvants
  - studying genetic variability of target antigens in the pathogen population, by using population genomics
  - studying the link between gene expression and protein formation
  - studying host-pathogen interactions, while focusing on antigen presentation on the one hand and the structure-function relationships of agonists and their host receptor on the other
  - understanding of the role of genes and proteins in complex immune responses or chronic disease progression
* defining criteria for choosing appropriate *in vivo* models:
  a) according to the pathogen of interest
  b) according to the research methodologies
  c) according to the industrial partners
* markers for monitoring the effectiveness of vaccine candidates
* better computational tools

* organized and centralized databanks

Legal needs

- create a legal framework enabling multi-antigen vaccines in terms of IP rights and pricing.
- create a legal and political framework enabling integration of different technologies and corresponding companies.

Conclusions

The world is currently facing an anti-infective crisis, stemming from various factors such as resistance to antibiotics, relative lack of biomarkers and the need for innovative treatments and drugs. The various "-omics" methodologies have the potential to boost Innovation, and their role will expand as they are increasingly used in anti-infective research, and in development of diagnostic and monitoring tools as well as drug targets and vaccines. Good diagnostic or monitoring tools permit a more informed use of antibiotics, improve pre-clinical tests for pharmacology and toxicology, aid in the identification and validation of possible biomarkers, and supply alternative endpoints to clinical ones, thus cutting down the costs of clinical trials.

Due to the increased demand for innovation, the need for coordinated, multidisciplinary research and political and legal efforts is more acute than ever before. Pharmaceuticals, diagnostics, therapeutic vaccines and biotechnology are likely to shift towards more collaborative business models. Therefore, the possibilities introduced to this field by genomics should be complemented by a substantive input from the different policy makers.

All "stakeholders", not only on the research front, but also the ones involved in licensing, administration, and financing therapy and health care in general, have a part to play in the development of more appropriate and standardised diagnostics, anti-infectives, vaccines and animal models. More rigorous evidence showing that new products either improve care or decrease further expenditures will likely be requested.

A more effective research and development process critically relies on the following points:

1) a political will to do so

2) a clear outline of the actual public health needs

3) better communication between the different partners

4) strengthening the financial background and bridging the development financing gap

5) fast track legislation for public health priorities

6) establishing an R&D educational program at the European level

There are some initiatives addressing existing problems, including efforts by PPPs, WHO, NIH, FDA, EMEA and regulatory authorities to obtain consensus solutions, but the battle is far from being won and continued efforts and initiatives are still needed.

Useful links:

- http://www.wipo.int/patentscope/en/patents.html World Intellectual Property Organisation
- http://lists.nottingham.ac.uk/mailman/listinfo/enemti ESF Network for Exchange of Microbial Typing Information (ENEMTI)
- http://www.escmid.org European Society of Clinical Microbiology and Infectious Diseases
- http://www.nccls.org/ Clinical and Laboratory Standards Institute
- http://www.europabio.org/ The European association for Bioindustries
- http://www.emea.eu.int/ European agency for the evaluation of Medical Products
- http://ec.europa.eu/enterprise/pharmaceuticals/index_en.htm European Commission Enterprise and Industry
- http://hivdb.stanford.edu/ HIV resistance database
- http://europa.eu.int/comm/enterprise/medical_devices/index_en.htm (Medical device sector for Industry sectors)
- http://www.pasteur.fr/recherche/genopole/PF8/betalact_en.html Genotyping of Pathogens and Public Health Platform
- http://www.lahey.org/studies/inc_webt.asp (attempt to standardize nomenclature of β-lactamases, and listing of available amino acid sequence variability)

http://www.c-path.org/ The Critical Path Institute (C-Path) is an independent, publicly funded, non-profit organization dedicated to the critical path initiative. C-Path fosters research and educational programs intended to enable the pharmaceutical industry to safely accelerate the development of new medications)

- http://www.GRACE_LRTI.org Scientific Network to tackle the increasing problem of antibiotic resistance in patients with chest infection. Stands for Genomics to combat Resistance against Antibiotics in Community-acquired LRTI in Europe.

Genome accession sites:

- The Institute for Genomic Research (TIGR): http://www.tigr.org
- Sanger Institute: ftp://ftp.sanger.ac.uk
- Stanford Genome technology Center: http://baggage.stanford.edu
- Stanford Univeristy: http://genome-www.stanford.edu
- Whitehead Institute: http://www-genome.wi.mit.edu
- Department of Energy Joint Genome Institute: http://www.jgi.doe.gov

Glossary

Proof of concept: A proof of concept is a short and/or incomplete realization (or synopsis) of a certain method or idea(s) to demonstrate its feasibility. The proof of concept is usually considered a milestone on the way of a fully functioning prototype.

Public Private Partnerships (**PPP**): This is a system in which a governmental service (academics), non-profit organizations, multilateral groups (WHO) and one or more private sector companies are linked by a partnership. In most existing health care related PPPs, capital investment is made by philanthropic foundations. Government contributions to a PPP may be the transfer of existing assets.

Cross licensing: The mutual sharing of patents between companies without an exchange of a license fee if both patent portfolios are about equal in value.

Pooled patents: Agreement between two or more patent owners to license one or more of their patents to on another or to third parties.

Venture Capital: Capital provided by outside investors for financing new, growing or struggling businesses. Venture capital investments generally are high risk investments but offer the potential for above average returns.

Business Angel or Angel Investor: An affluent individual who provides capital for a business start-up, usually in exchange for ownership equity. Unlike venture capitalists, angels typically do not manage the pooled money of others in a professionally-managed fund. However, angel investors often organize themselves into **angel networks** or **angel groups** to share research and pool their own investment capital. In

start-up financing, Angel capital fills the gap between the "three F"s (friends, family and fools) and venture capital.

Exclusive license: a single licensee has the right to use the patented technology, which cannot be used even by the original patent owner.

Sole license: a single licensee and the patent owner have the right to use the patented technology.

Non-exclusive license: several licensees and the patent owner have the right to use the patented technology.

Sepsis: Spectrum of clinical conditions caused by an immune response to infection that is characterized by inflammation and coagulation.

Synthetic lethal screen: Screening method used to uncover mutations in a second gene that will require the cell to maintain a wild-type copy of the gene being studied in order to survive.

Innate immunity: Immunity that is naturally present and is not due to prior sensitization to an antigen. Since it is not stimulated by specific antigens, innate immunity is generally non-specific.

Prophylactic vaccine: Preventive vaccine which protects against an initial microorganism infection.

Therapeutic vaccine: Vaccine which helps those already infected by a microorganism by enhancing and strengthening the immune response against the microorganism. These vaccines selectively stimulate the immune system against specific infections.

Acronyms

CEO	Chief Executive Officer
ECDC	European Centre for Disease Control
EMEA	European Agency for the Evaluation of Medicinal Products
ESBL	Extended spectrum producing beta-lactamases producing strains
FDA	Food and Drug Administration (US)
IP	Intellectual Property
MOA	Mode of Action
MRC	Medical Research Council (UK)

MRSA	Methicilin-resistant *Staphylococcus aureus*
NIH	National Institutes for Health (US)
PPP	Public Private Partnerships
R&D	Research and Development
SME	Small and Medium Enterprises
SNP	Single Nucleotide Polymorphism
TB	Tuberculosis
TLR	Toll-like receptor
VC	Venture Capitalist
VRE	Vancomycin-resistant enterococcus
WHO	World Health Organisation

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