

MEDICAL BIOTECHNOLOGY - MODERN DEVELOPMENT

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Current products and processes of medical biotechnology have already revolutionized the practice of medicine in terms of better diagnostics and therapeutics, but the more advanced ideas and products have not yet been brought into clinical practice. It is important to understand that multiple factors have contributed to its development and growth, including concurrent advances in computational science, cell technology, and a measure of serendipity. On a global perspective, medical biotechnology is recognized for its potential to stimulate the economy as well as reduce health inequities. However, this will not happen by itself. Left on its own, market forces will result in a tendency for health inequities to widen in developing countries, and diseases afflicting the poor to be neglected. A considerable degree of organization, regulation, and careful policymaking and implementation are required for the optimal application of medical biotechnology and reduction of health inequities. As with all new innovative technologies, a number of social and ethical issues are raised with each new development. Some of these, such as opposition to stem cell research and genetic databases, will be reduced over time with education and familiarity. Others, such as reproductive cloning, may never be accepted. Regulations and constant dialogues between scientists, policymakers and the lay community are necessary to minimize the misuse of medical biotechnology and assuage public anxiety. The future of medical biotechnology is very bright.

1. Introduction

Medical biotechnology, also termed red biotechnology, is the application of biological techniques to product research and development in healthcare and medicine. Breakthroughs in this and associated scientific fields have revolutionized the practice of medicine: newer and simpler tests for the more accurate diagnosis of disease; genetic and proteomic tests that allow for prevention of disease; more efficient methods for designing and making drugs that are targeted at the molecular level and therefore conceivably more effective but less toxic; the possibility of gene therapy to cure diseases that are previously incurable. The fulfillment of the vision of “individualized medicine”, where therapy can be tailored to disease of the individual seems to draw closer with each passing day.

Although genomics and its applications (viz. gene therapy) commonly come to mind when medical biotechnology is mentioned, it should be remembered that other disciplines such as bioinformatics (see also *Bioinformatics on Post Genomic Era- From Genomes to Systems Biology*), nanotechnology (see also *Nanomedicine and Nanorobotics*), fermentation technology (see also *Basic Strategies of Cell Metabolism*) and cell technology (see also *Microbial Cell Culture*) also play an important role in the

development of the field. Nevertheless, the golden era of medical biotechnology and indeed, of biotechnology in general, began only in the 1970's with the development of recombinant DNA (rDNA) techniques (see also *Methods in Gene Engineering*). The initial experiments of Paul Berg and his team at Stanford University in 1972, where the genome of SV40 – a simian virus – was attached to a segment of DNA in a common bacterium, led to the first rDNA produced. Their gene-splicing techniques, which were improved upon by others, allowed for genes coding for different proteins to be inserted into foreign cells. The recipient cells could then be induced to produce the desired proteins. Such techniques set the foundation for a whole array of biologic research, including the subsequent sequencing of the human genome.

The great potential of biotechnology for wealth creation and economic growth did not escape notice, and many countries – mostly developed countries – and multinational companies have made huge investments into research and development (R&D) of life sciences and biotechnology. However, the returns have been less than impressive on a worldwide scale for public biotechnology companies. Ernst & Young, currently one of the largest professional service firms worldwide, reported an industry net loss of USD 5.4 billion in 2006 despite total revenues of USD 73.5 billion – this figure is a 35% increase over the net loss of USD 4.0 billion in 2005. One caveat of this and other biotechnology industry reports is that it is difficult to separate out the medical biotechnology companies from the rest of the industry, with the exception of pharmaceutical companies. Nevertheless, the industry as a whole continues to grow, as more countries inject more capital into biotechnology R&D each year.

On a social front, it is important to understand that welcome for the products of medical biotechnology has not been universal. Significant professional, religious and public reservation remains on the potential abuse of genetic information, risks of therapy, and ethics of research. It is also clear that the financial costs of these products can be considerable, and this can widen the already significant gap between the healthcare options of wealthy individuals and developed countries, and the poorer individuals and nations. However, the potential of medical biotechnology to contribute to improving human health and wellbeing cannot be denied – the United Nations in their Human Development Report in 2001 viewed biotechnology as one of the most important means of dealing with the expanding needs and major health challenges faced by poor developing countries.

2. Diagnostics

It is in the area of diagnostics where biotechnology has arguably been most successful from the point of view of transforming practice, although the market for diagnostic products is considerably smaller than the market for therapeutics. The two major contributions of medical biotechnology to diagnostics presently, beyond improvements over the sensitivity and specificity of conventional tests, are:

- Pre-diagnosis – the ability to screen for and detect the predisposition for diseases in individuals, and
- Prognostication – better prediction of outcomes for particular diseases, or the effects of therapy on the patients

The majority of successful commercial diagnostic tests introduced as a consequence of medical biotechnology advances are nucleic acid tests and those tests based on monoclonal antibodies. Considerable effort has also been expended to harness the technologies used for proteomics research for the purpose of advancing medical diagnostics. As with the application of nanotechnology to diagnostics (i.e. “nanodiagnostics”), however, these efforts have not yet yielded results or applications that can be widely utilized.

2.1 Nucleic Acid Tests

The fundamentals of genetic replication, first discovered in 1953 by Watson and Crick, laid the foundations of clinical molecular diagnostics, but it was only 22 years later that Edwin Southern’s DNA hybridization technique (Southern blot) brought it into being. This was a labor- and time-intensive technique, however, and limited molecular diagnostic testing to low-volume work in large institutions. It took a further development – the invention of the polymerase chain reaction (PCR) in 1983 by Kary Mullis – to revolutionize molecular diagnostics.

The PCR is an in-vitro technique for isolating and exponentially amplifying a fragment of DNA via enzymatic replication (see also *Chemical Methods applied to Biotechnology*; and *Physical Methods applied to Biotechnology*), and its ingenuity lies in its capability of amplifying even trace amounts of specific DNA to detectable levels. In its current iterations, it is commonly performed via semi-automated instruments that may be entirely automated at high costs via front-end robotics. The coupling of DNA amplification with fluorescence-based detection results in what is known as “real-time PCR” – wherein quantification of DNA occurs at the end of every amplification cycle to give highly sensitive and specific yet rapid results. Modern molecular diagnostics is currently still based predominantly on PCR, and coupled with expanding knowledge and databases of human and pathogen genomics (see also *Human Genetic Databanks : From Consent to Commercialization - An Overview of Current Concerns and Coundrums*), has led to it becoming the cornerstone for the diagnostic work-up of an ever increasing list of diseases. Some examples are given below.

2.1.1. Role in Infectious Diseases

Nucleic acid tests for the human immunodeficiency virus (HIV) can quantify the amount of HIV in a patient’s blood sample (see also *Blood: The Essence of Humanity*). Although seldom used for the diagnosis of HIV in adults because of their relative cost, they can be used to test infants born to HIV-positive mothers – even if uninfected, these infants may have a positive HIV antibody test result because they carry maternal antibodies within their circulation. However, the major uses for these molecular tests are in the screening of donated blood in blood banks – to reduce the so-called “window period” where HIV may be undetectable in newly-infected patients – and as a way to evaluate the success of anti-retroviral therapy by measuring the degree of reduction of the HIV viral load in treated patients. Genotypic assays, to determine the presence of drug resistance determinants in HIV are also available, and recommendations about their use are now incorporated into the HIV treatment guidelines in most developed

countries, such as the Department of Health and Human Services guidelines in the US.

Similar tests to quantify the amount of virus in infected patients are available for Hepatitis B and C viruses, cytomegalovirus, and Epstein-Barr virus among others. As with HIV nucleic acid tests, these are used less for disease diagnosis in patients – there are cheaper and equally reliable antibody tests for diagnosis – than for prognostic determination and evaluation of the effects of therapy.

In diagnostic testing for bacterial antibiotic resistance, PCR-based assays for the detection of methicillin-resistant *Staphylococcus aureus* (MRSA) – a Gram-positive bacterium responsible for considerable mortality and morbidity in the hospitals of most developed countries – directly from patient samples have reduced the time to diagnosis from 48 to 96 hours using conventional culture techniques to just two to four hours. The majority of these tests depend on the intrinsic capability of the PCR for amplifying minute amounts of DNA to detect for the presence of the *mecA* gene responsible for drug resistance from the small numbers of bacteria potentially present on patient samples. From the economic and public health perspectives, the expense incurred to isolate and contain potentially communicable diseases far outweighs the increased costs of such novel diagnostic tests.

2.1.2. Role in Cancer

For hereditary cancers, genetic screening using PCR-based methods combined with bioinformatics databases and disease registries offers the possibility of earlier diagnosis or even pre-emptive therapy. The classic example is colorectal cancer associated with familial adenomatous polyposis (FAP). Commonly caused by mutations in the tumor-suppressor *APC* gene on chromosome 5, this condition is inherited in an autosomal dominant fashion, and results in colorectal cancers developing before the age of 40 years in affected individuals. Genetic testing allows for the identification of family members who possess the gene, and who are thus at extremely high risk of developing cancer. These individuals can then be targeted for counseling, regular colonoscopies and even prophylactic surgical removal of their colon if extensive polyps – pre-tumorous lesions – develop. Other examples include the identification of *BRCA1* and *BRCA2* that predisposes females to early onset breast and ovarian cancers. The ability to identify *BRCA* carriers has allowed counseling of at-risk families, earlier diagnosis and prophylaxis of these cancers.

Genetic testing has also allowed for the identification of particular genetic mutations in cancer with prognostic significance. Examples include the recent identification of epidermal growth factor receptor (EGFR) mutations in patients with non-small cell lung cancer. Patients harboring sensitizing EGFR mutations have a more benign disease course compared to others. The identification of the *c-ret* oncogene in medullary thyroid cancers may also identify existing cancer patients who have genetic predispositions to familial cancer syndromes, thereby allowing for early diagnosis of other cancers and associated conditions in the index patient.

2.1.3. Prenatal Screening and Pre-implantation Genetic Diagnosis

Prenatal genetic screening is offered in most developed countries, with the rationale of detecting chromosomally abnormal conceptions and the presence of genetic disorders, and thereby offering the option of terminating conceptions early. Common conditions screened using nucleic acid tests include Down's syndrome and other chromosomal aberrations, hemophilia, cystic fibrosis, etc – the list expands with commercial availability. As with genetic screening in general, this remains a contentious area with legal and ethical implications.

Pre-implantation genetic diagnosis (PGD), i.e. genetic diagnostic tests performed on embryos prior to implantation, is an alternative to prenatal diagnosis. It followed the success of in-vitro fertilization (IVF) procedures, with first successful attempts at human testing performed in 1989 by Handyside and co-workers, where PCR was used for sex determination in the embryos of patients with X-linked diseases.

2.2 Monoclonal Antibodies

Georges Köhler, César Milstein, and Niels Kaj Jerne won the Nobel Prize in Physiology or Medicine for their discovery of the process of producing monoclonal antibodies in 1975. By fusing specific antibody-producing cells with myeloma cells that had lost the ability to produce antibodies, researchers could create hybrid cells (hybridomas) that would produce identical (i.e. monoclonal) antibodies that could theoretically hone in and bind to any given substance – a literal fulfillment of Paul Ehrlich's "magic bullets".

The development of monoclonal antibodies was a minor revolution in healthcare, and their use rapidly increased in both diagnostics and therapeutics. In the field of pathology, they are now routinely used as part of immunohistochemistry to detect antigen in fixed tissue sections, thus facilitating diagnosis. Tagged with radio-isotopes and injected into patients, monoclonal antibodies can improve the precision of surgery by pinpointing the location of target cells. George Stark's western blot technique for detecting proteins, perhaps best known for its use in HIV confirmatory testing, is also dependent on monoclonal antibodies directed against the target proteins. The role of monoclonal antibodies in therapeutics will be described in a later section.

2.3. Proteomics for Diagnostics

The earliest attempts at using proteomics for clinical diagnostics involved using high-throughput investigations to identify novel disease biomarkers via surveying the clinical samples of healthy and diseased individuals. Proteins found to be differentially distributed between these samples were then selected with a view of identifying them as potential biomarkers. However, this strategy has not been successful to date possibly because of technical issues, i.e. current technologies are incapable of detecting low protein concentrations; issues related to inter-individual variability, i.e. protein level differences related to age and gender; and issues related to the disease, i.e. most diseases may not have one single unique biomarker.

Recent attempts have focused on the identification of protein pattern signatures, using a technology termed single emission laser desorption ionization time-of-flight mass spectrometry (SELDI-TOF-MS). Simply put, biological fluids of interest are applied to

an array surface and ionized, and the desorbed proteins' mass:charge ratios are then measured via TOF-MS. Powerful computational and bioinformatics programs then derive diagnostic patterns from each profile. Lance Liotta's team first described the use of SELDI-TOF-MS in diagnosing ovarian cancer in 2002, and since then, there have been multiple publications on its use in a variety of clinical diseases. However, although potentially revolutionary, this technology suffers from the same issues as Southern blot in the 1980's – high operational costs and operator expertise requirements have limited its use to only a few large regional institutes worldwide at this point in time.

2.4. Nanodiagnostics

The application of nanotechnology in clinical diagnostics is relatively new, even compared to proteomics. Rather than searching for new biomarkers, as is the case for much of biotechnology research in diagnostics, research in nanodiagnostics is mainly centered on extending the limits of current diagnostic techniques. A prime example of this is research in microfluidic or “lab on a chip” systems, with the idea of combining the numerous processes of DNA analysis onto a single glass and silicon chip. Within a chip the size of a conventional microscope slide are fluidic channels, heaters, and all the devices present within considerably larger PCR machines. Another example is the “pill-camera” used to detect gastrointestinal bleeding, powered by microelectromechanical systems (MEMS). Within a capsule the size of a regular tablet are a video camera, optics, a light-emitting diode, and a transistor. Images taken by the camera are transmitted to an external computer for analysis. This obviates the need for more risky gastrointestinal endoscopy.

Although nanotechnology opens up a wide array of possibilities in the future of clinical diagnostics, there are no mass-market products available at this point in time. It remains to be seen if its great promise can be fulfilled in the near future.

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Biographical Sketches

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