

Pharmacogenetics Workshop
Background paper
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Friday 29th October, 1999
The White House Hotel
London



The Wellcome Trust

Introduction to this Paper and the Workshop

This paper is intended as a background briefing document to provide an overview of the field for participants in the Wellcome Trust's workshop on pharmacogenetics. The Trust commissioned the paper from science writer, Robert Snedden. It is not intended to provide a definitive statement of either the scientific or of the social, ethical and public policy issues arising from developments in pharmacogenetics.

The purposes of this paper are to provide a basic introduction to the science for those who may not be intimately familiar with the area, and to raise some of the questions which might be generated by new developments in pharmacogenetics.

Answering these questions will require research and analysis. It is hoped that the workshop will prompt participants to think of more research questions and to devise appropriate research methodologies. The workshop is not intended to debate issues, but rather to start asking research questions. Ultimately, the hope is that the results of research and analysis might feed into rational public policymaking.

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PHARMACOGENETICS

Ninety-nine point nine per cent of all human DNA is identical from one person to the next. The one-tenth of a per cent that is unique plays its part, along with environmental influences, in shaping the differences between us in outward appearance and in the way we act. Our genes also play a role in the ways in which our bodies vary in their response to the disease agents we encounter and in their ability to metabolise the medicines used to treat those diseases. The variability among humans that is so apparent in the obvious features such as height, skin colour and susceptibility to disease is true also of drug metabolism.

In the beginning

Reasoning from his experience with alkaptonuria (black urine), the English physician and molecular biologist Archibald Garrod developed the concept of biochemical individuality at the beginning of the twentieth century. Taking his lead from the newly rediscovered work of Gregor Mendel, Garrod reasoned that alkaptonuria sufferers were carriers of a Mendelian recessive characteristic. He called them 'chemical sports'. In his book *Inborn Errors of Metabolism* (1909) he described several diseases that were caused by genetic errors.

A case of porphyria brought on by ingestion of the hypnotic drug sulfonal led him to implicate enzymes in drug detoxification. He suggested that the adverse effects that some substances had on certain individuals were due to failure of the enzymes to detoxify them. Garrod spoke of 'Inborn errors of metabolism', which were 'due to the failure of a step in the metabolic sequence due to loss or malfunction of an enzyme.' This was the first step, albeit unknowingly, towards the science of pharmacogenetics. It would be several decades before researchers began to link human chemical variation with their genetic make up.

In 1941 George Beadle and Edward Tatum deduced from their studies of mutations in the bread mould *Neurospora crassa* that the formation of each individual enzyme is controlled by a single, specific gene. This one gene, one enzyme hypothesis found wide applications in biology and marked the beginning of the science of biochemical genetics.

The study of genetically controlled variations in the response to drugs is still a relatively new field of scientific inquiry. Ground-breaking studies of the therapeutic agents succinyl-choline, primaquine and isoniazid in the 1950s marked the true start of pharmacogenetics as an experimental science. Following the introduction of succinyl-choline as a muscle relaxant during surgery, for example, variations in patients' responses to it became apparent. The effects of succinyl-choline are generally short lived, but 1 in 2000 people are unusually sensitive to it. Succinyl-choline is usually metabolised by serum cholinesterase, however some individuals carry a variant serum cholinesterase with reduced activity and the drug takes longer to degrade, which means that its effects are longer lasting.

In 1957, Arno Motulsky in an article printed in the *Journal of the American Medical Association*: 'Drug reactions, enzymes and biochemical genetics', was the first to suggest that genetically determined variations in the activity of drug-metabolising enzymes in the liver were the cause of certain adverse drug reactions. Two years later, in 1959, the term 'pharmacogenetics' was first proposed to describe the relationship between genetic make up and drug response. Motulsky is currently carrying out investigations in ecogenetics, studying the role of genetic polymorphisms in the resistance and susceptibility to disease from environmental agents, at the University of Washington. (Some people would classify pharmacogenetics as a subclass of ecogenetics.)

The revolution in molecular biology that began when Watson and Crick worked out the structure of DNA has taken off at the end of the twentieth century with the advent of

more and more powerful tools for genetic analysis. Among the changes that many observers are expecting in the twenty-first century is a shift towards disease prevention and treatment using individually tailored pharmaceuticals that manipulate genes and cells.

By the beginning of 1999 Randall Tobias, chairman emeritus of Eli Lilly, could announce, perhaps rather optimistically, at the New York Academy of Sciences that: 'Something truly amazing is occurring. Medicine is today about where aerospace was in 1927 when Lindbergh flew across the Atlantic. In the not-so distant-future, though, the life sciences will have accomplished the biology equivalent of putting a man on the moon. The day will come when we regard all surgeries, except trauma, as failures of the pharmaceutical industry.'

What is pharmacogenetics?

Pharmacogenetics brings together pharmacology (the study of how drugs work in the body) and genetics (the study of how characteristics are inherited). Pharmacogenetics is the study of the way in which an individual's genotype, their genetic make up, can influence their response to drugs. It can help to address the problem of why some patients respond well to drugs and others do not. It can also help doctors understand why some patients require higher or lower doses of a particular drug. The goal of pharmacogenetics research is to provide information for the practise of 'personalised medicine' – getting the right medicine, at the right dosage, to the patient.

A great deal of pharmacogenetics research has focused on the mechanisms that determine drug concentration within the body, looking at the end result of ingestion, absorption, metabolism, clearance and excretion of a drug. Genetic tests are being developed to determine a patient's ability to metabolise a particular drug, which will

allow dosage to be determined with greater certainty, and also to determine which patients may be susceptible to adverse side effects as a result of being prescribed a drug. The pharmacological consequences of genetic variation are highly diverse. The absence of drug-metabolising enzymes can cause excessive, and fatal, drug action, or the therapy may fail because the drug is not activated. For example, codeine fails to cause analgesia if it is not converted to morphine by enzyme action – a condition that has been found in 10 per cent of white Americans.

Pharmacogenetics versus pharmacogenomics

Classical pharmacogenetics depended on the observation of phenotypes, the visible manifestation of the combination of genotype and the environment, particularly relating to drug metabolism. The expanded field of enquiry that has been opened up by the availability of molecular techniques is now often referred to as pharmacogenomics. Pharmacogenomics represents the merging of pharmacogenetics with the new technologies of genomics. Pharmacogenomics is concerned with the discovery of new drug response genes and the development of novel molecules to target these genes, indeed pharmacogenomics has been used by some commentators to refer to the commercial application of genomic technology by the pharmaceutical industry. Understanding the genetic factors responsible for a patient's response to a drug makes it possible to design predictive tests before exposure to the drug. Pharmacogenomics involves both developing drugs that will target a particular disease and tailoring them to take into account a person's genetic make up so as to avoid any side effects that might be associated with their use. The terms pharmacogenetics and pharmacogenomics are often used interchangeably to mean much the same thing (this paper is probably guilty of doing just that).

The knowledge of human genetics that will become available as the Human Genome Project progresses will be invaluable in developing pharmacogenetics. By spring next year, researchers will have produced a rough draft of practically all of the human

genome. The final, finished sequence will be available three years after that.

Unravelling the function of each gene will take considerably longer but as more information is uncovered pharmaceutical companies will be able to put genetic data to use in developing a new generation of therapeutic drugs. Drug design will rely less on trial-and error-clinical trials involving real people and all the messy ethical considerations that involves. Instead pharmacists will be able to create new classes of medicines based on gene sequence and information on the structure and function of the proteins the genes code for. Medicines like this, tailored to the specific genome of an individual, should have fewer side effects than many of the currently available medicines.

Pharsight, one of the new breed of pharmaceutical companies based in Palo Alto, California, conducts clinical trials that involve no human subjects at all. Stuart Koretz, Pharsight's vice-president of medical affairs, has suggested that between a quarter and half of all trials make no meaningful contribution to the data on a drug candidate. By running various computerised versions of trials, each with different assumptions, the regimes that prove useless can be avoided in the real trials that follow the computerised ones. Data on attributes such as toxicity, pharmacokinetics, pharmacodynamics and what is known about the behaviour of any similar drugs, are used to program large populations of software 'volunteers' with a variety of responses which, Pharsight hopes, will accurately reflect potential patients in the real world.

Pharmacogenetics Research

Traditionally, pharmacokinetics and pharmacodynamics have described how drugs affect the physiology of the body, and they have been used to establish the dosing for any given drug. The ultimate goal of pharmacogenomics is to produce a genetic profile of the patient that reflects pharmacokinetic and pharmacodynamic parameters, and which can then be used in a diagnostic and therapeutic assessment.

However, a great deal of work must be done before this goal can be achieved.

Historically, an inherited variation only became apparent as a result of the clinical observation of an unusual response to drug therapy in an individual. In recent years advances in molecular biology and genomics have brought further discoveries in the field of pharmacogenetics.

Specific genotypic information will provide physicians with the means to select the optimum therapy to achieve the desired clinical outcome. Links between genotype and drug effect are not yet definitely established, especially since, in many instances the observed effects may be due to multiple gene action. Before genetic profiling becomes an established part of clinical procedure, carefully designed trials incorporating genetic analysis will have to be carried out in order to establish the predictive value of genotyping with regard to drug response. At an American Medical Association media briefing on genetics in June this year Ira Herskowitz, Professor of Genetics, University of California San Francisco, said 'With a better definition of disease and how an individual responds to a particular candidate drug, drug trials can be carried out on just the right people who will metabolise the drug properly. This, therefore, greatly increases the chances of having a successful outcome for a clinical trial.'

Drugs are designed and prescribed by and large to suit a generalised patient population. The way in which a particular patient will react to a drug may be dependent on a number of factors and these will include environmental as well as genetic influences. Insofar as these differences in response can be attributed to genetic variation between individuals, however, it is the aim of the pharmaceutical industry to find the means to control for them, not only when a drug is prescribed, but also when it is tested. Axys Pharmaceuticals in La Jolla, California, has been focusing much of its research into the study of computer models of trials of drugs metabolised by the cytochrome-P450 complex of hepatic enzymes. P450 variations have long been recognised as an important source of the differences in people's responses to drugs. Janice Kurth, Axys

director of pharmacogenetics, estimated that data on the complex could allow the size of clinical trials of certain cytochrome-sensitive drugs to be cut by 85 per cent

Adverse reactions

As George Poste, Chief Science and Technology Officer for SmithKline Beecham, puts it in the October 1998 edition of *Nature Biotechnology* when a patient is given a drug 'everything is a crap shoot, and you hope that the adverse reactions [to the drug] aren't too great based on the empirical therapy that is practised on you.'

In the same article Poste quotes a study in the *Journal of the American Medical Association* (15 April, 1998) showing that two million Americans suffer from adverse drug reactions every year. Of that number '106,000 don't leave the hospital because they die'. This makes adverse drug reaction the fourth largest cause of death in the United States. At Bio 98, the Biotechnology Industry Association's international meeting in New York in June 1998 Poste stated that up to 35 per cent of people do not respond to beta blockers, and as many as 50 per cent do not respond to tricyclic antidepressants.

Dr Daniel Cohen, chief genomics officer of Genset in Paris, also speaking at Bio 98, said that 85 per cent of a patient response to drugs is due to genetics. 'Patients may have to wait several months, even a year, before they know if a drug works. They may have to try several drugs one after another. We would like to be able to say: "This drug will be good for you."' Clinical trials comparing the genes of patients who respond to a drug with those who do not could lead to the development of a test that would identify those who should get the drug.

Prescribing and regulating drug dosage will no doubt become more sophisticated. The present methods of determining dosage based on weight and age will be replaced by a more sophisticated tailoring of drug dosage to the patient's genetically determined

ability to metabolise the drug. For example, a patient who does not produce very much of a particular metabolic enzyme can be prescribed lower doses of the drugs that interact with that enzyme.

Another factor for pharmaceutical companies to take into account is that of liability. Companies may find themselves obliged to market tests that will determine who will benefit or, perhaps more importantly, who may be harmed by a particular drug. A company marketing a drug that showed adverse reactions in some of those patients for whom it was prescribed could face litigation if it could be shown to have been negligent in carrying out clinical trials to establish responses based on genetic variation. Each one of George Poste's two million hospitalised Americans represents a possible legal action. 'Imagine,' he says, 'a lawyer asking, "Doctor, did you know this drug would kill your patient? Did you know there is a test that would have predicted that? And why did you not give your patient that test?"'

The genetics of drug response

There are several reasons why individuals may respond negatively or not at all to a particular drug. In general most variations in response to a drug have been found to be due to variations in a specific gene. Individual variations in genes that code for proteins in a metabolic pathway can affect the metabolism of a drug and thus its effectiveness. Drug reactions may include hepatotoxicity, cardiovascular problems, or blood cell reactions or they could be reactions caused by interaction with other drugs.

The importance of individual variation in the response to treatment has been widely accepted since the nineteenth century. In the past this variation has been managed by careful descriptions of potential side effects with clinical information on drug metabolism being obtained by the administration of bioactive compounds to a trial population for the sole purpose of determining the relative rate of metabolism. By

doing so clinical trial populations could be divided into subsets according to metabolic rate, and metabolically associated adverse reactions could be avoided.

For the pharmaceutical industry an obvious motivation for pharmacogenetic research will be in the cost savings that could come from testing for genetic variations. One of the first such variations to be widely tested for could be one that affects a person's response to asthma drugs. Beta-two agonists, which include Ventolin and Albuterol, act on the smooth muscle of the lungs that control the opening and closing of the bronchial tubes, which are constricted in asthma. The target protein in the smooth muscle comes in at least two variants and response to the asthma drug will depend on which variant the patient has. The beta-two agonist has little effect on one variant, which makes the treatment next to useless in such instances.

As more and more of the genes that influence drug response are discovered, there is likely to be an increase not only in the number of tests carried out, but also a difference in the way these tests will be used. Panels of tests that might cover a variety of different drugs all related to a single disease could be carried out simultaneously. With such a panel, it should be possible not merely to give the go ahead to a specific treatment but to select from among many available therapies the one that has the greatest potential benefit.

It can take more than £220 million and fifteen years to develop a new drug, and between 80 and 90 per cent of the drugs entered for clinical trials never even make it on to the market. When they do get to the pharmacy they may be ineffective for many people. Three quarters of all patients on Tagamet, the world's best-selling anti-ulcer drug, experience little or no clinical benefit, for example. Procainamide, a drug used to treat heart arrhythmias, can cause a potentially fatal liver disease in people whose genes make them slow to carry out a certain biochemical reaction.

Dr Fred Ledley, president and CEO of Variagenics in Cambridge, Massachusetts, said: 'Ultimately economic issues will drive pharmacogenomics into conventional practice: the cost of drug failures – drugs given to people that don't work, that delay proper treatment, that increase the cost of care, the expenses of treatment toxicity, and the expense of return visits.' The pharmacogenetic approach may allow pharmaceutical companies to look again at drugs that have failed clinical trials because of low response rates in the general population, targeting them at the people for whom they are suited.

The other side of the coin is that personal pharmaceuticals may not prove popular with big pharmaceutical companies, which need to offset the huge development costs involved in bringing a new drug on to the market. Pharmacogenetics is about developing drugs that are targeted very specifically, which means that they should be both effective and safe. However, the very fact that pharmacogenetic drugs will be targeted to smaller markets than traditionally developed drugs is seen as a drawback. The pharmacogenetic strategy will only work if the drug companies can charge a high enough price for their product. Of course as Dr William Evans, chairman of the pharmaceutical sciences department at St Jude's Children's Research Hospital pointed out recently, 'Pharmaceutical companies may develop a drug that only 10 per cent of the market can use, but 100 per cent of that 10 per cent will use it.'

Healthcare economics

It is not only the pharmaceutical companies that are concerned with the cost-benefit analysis of implementing pharmacogenetic solutions in health care. It would be for the health care provider, whether National Health Service or private health insurers such as the HMOs in the United States to find the money to pay for potentially expensive treatments. The ideal situation for the health providers is that the medical service being implemented should be high volume and low cost. If the technology of genetic screening and analysis becomes sufficiently automated this ideal may be approached for pharmacogenetics. However, if pharmacogenetics falls in the high volume/high cost

side of the equation then there will be concerns from health care managers over the justification for its use.

Another point to consider is the possible advantage to be gained by private health care organisations over their competitors if they are the first to offer tailored health care to their customers. The current 'trial and error' method of the physician prescribing a 'best guess' treatment for the ailment being presented can itself be a lengthy and potentially expensive exercise if the patient re-presents because the initial choice has proven to be ineffective. (Discussions of patient psychology and the importance of the placebo effect in medical treatment are perhaps outwith the parameters of this paper but will doubtless suggest themselves as matters to be considered by many readers.)

Cholesterol-lowering drugs, for example, are one case of an expensive treatment that is ineffective in non-metabolisers of the drugs. Pharmacogenetics could prove cost-effective if it enables doctors to identify non-responders straightaway without waiting three or four months before trying another drug.

Orphan Drugs

For a third of people with rare diseases it can take from one to five years to get a diagnosis and for 15 per cent it can take more than five years. Once diagnosed, however, they may find there is little they can do about it. For some, in the absence of any other form of treatment, there may be the possibility of clinical trials of experimental drugs. Even when drugs do exist for the treatment of a rare condition, they can be expensive, and insurers may refuse to pay for them. Because the potential market for many drugs is tiny, companies often charge a lot. The 2,500 patients in the United States taking the drugs Ceredase and Cerezyme for Gaucher's disease, a rare disorder of lipid metabolism, face costs of \$150,000 to \$170,000 a year, for example.

The Orphan Drug Act, passed in the United States in 1982, defines an orphan disease as a condition that affects fewer than 200,000 persons in the United States. According to the National Organisation for Rare Disorders more than 20 million Americans suffer from one or other of 5,000 of these rare conditions. Because no pharmaceutical company was prepared to meet the cost of developing or adopting the products necessary to treat these illnesses they became known as orphans.

Until the Orphan Drug Act was passed, patients suffering from diseases such as multiple sclerosis, haemophilia, Parkinson's disease and some rare cancers found themselves without adequate health care. Without a large target population there was no financial incentive for the pharmaceutical companies. Even so, there were a few instances of pharmaceutical companies developing drugs with limited commercial potential in the 1960s and 1970s such as Mithracin (plicamycin), which is used in the treatment of testicular cancer.

In 1982 legislation was passed by the US Congress that offered generous incentives to companies that were willing to adopt orphans and develop and market treatments for them. Since the act came into force the FDA has approved more than 180 orphan products. Over 1200 applications have been made for orphan designation, of which the FDA has approved over 900. Being granted orphan designation allows a company to proceed with development and take advantage of the law's financial incentives.

The Orphan Products Grant Program managed by the FDA awards up to \$200,000 a year for a maximum of three years to those engaged in clinical trials on the safety and effectiveness of orphan products. Close to 400 studies have been funded so far. In addition, sponsors may claim 50 per cent of their clinical trial costs as a credit against taxes.

To take one example, in 1998 the FDA cleared thalidomide, still a highly controversial drug because of the birth defects caused by its use by pregnant women in the 1960s, for

the treatment of Hansen's disease (leprosy). Thalidomide can be used to treat a serious inflammatory symptom seen in Hansen's patients. Because of its reputation severe restrictions were put on the drug's use. Before the Orphan Drug Act no one would have carried out the research that uncovered this therapeutic use for thalidomide. The drug has also been granted orphan designation for the treatment of primary brain tumours and Kaposi's sarcoma, an AIDS-related cancer, although it has not yet been approved for this use.

Only about 15 per cent of applications for an orphan designation come from the large pharmaceutical companies. What is happening is that smaller concerns are being formed specifically to take advantage of the legislation and develop and market orphan products for a tightly focused market. John McCormick, deputy director of the FDA's Office of Orphan Products Development, believes that the orphan law can be given a lot of the credit for the establishment of the American biotechnology industry. He points out that the law grants seven years of exclusive marketing rights for orphan products and that this allows a small company to bring its product to market without worrying that a larger concern might bring out a competing product.

The European Commission put forward a proposal in July 1998 in order to establish procedures and measures aimed at encouraging the production and market of orphan drugs within the European Union. It did so on the grounds that a common Community approach was more likely than isolated national initiatives to encourage the pharmaceutical industry to develop products for which there would probably be a limited demand, given the high cost of doing so. The range of measures contemplated include the granting of market exclusivity for ten years and the proposed waiver of the evaluation fees normally charged by the European Agency for the Evaluation of Medicinal Products (EMEA). The British Government supported this initiative in principle, seeing it as reasonable for the Community to bear the cost of orphan drugs regulation rather than having any shortfall met by higher fees on non-orphan products. A Regulatory Impact Assessment suggested that the main cost to the British

Government would come from its contribution to the Community subsidy to the EMEA. This has been put at around £687,000 over the four years 2000-2003.

Orphan drugs and pharmacogenetics

As more companies take a pharmacogenetic approach to drug development a dramatic increase in requests for orphan drug status is likely. Herceptin would seem to qualify without question as an orphan drug. Discovered by Genentech it was approved by the FDA in 1998 for the treatment of metastatic breast cancer. Herceptin is a monoclonal antibody directed against the HER2/neu protein (a cell-surface receptor) that is present in all normal tissue and overexpressed in some breast tumours. It is presumed that Herceptin will only be effective in patients who are HER2/neu overexpressers, therefore before a woman can be prescribed Herceptin, her tumour must be tested to ensure that it falls within this category. An estimated 30 per cent of all breast cancer patients have tumours of this kind.

There are roughly 165,000 women in the United States with metastatic breast cancer. Clearly, 30 per cent of this number falls some way beneath the FDA definition of an orphan disease affecting under 200,000 people. Even so, Herceptin was denied the orphan-drug designation.

The Office of Orphan Products Development (OOPD) defines a drug's patient population as the total expected treatment population, not just those whom the pharmaceutical company identifies as eligible for clinical trials. Products are most commonly denied orphan-drug status because of disagreements over how the target population is defined. The pharmaceutical companies are not above dividing markets up creatively so that they can claim fewer than 200,000 patients. For example, companies will conduct trials only with those patients for whom standard therapy has failed; thus ensuring that the drug is approved solely for use with these patients.

However, the OOPD would include the entire potential patient population as there is no reason, why the drug could not be used as a first choice therapy.

In the case of Herceptin (the OOPD do not discuss the reasons for their decision) the question may have arisen as to whether Herceptin should legitimately be restricted to metastatic patients as HER2/neu is overexpressed in other forms of cancer as well as breast cancer. The enlarged potential patient population thus created may have been the reason for the withholding of the orphan-drug designation.

In fact, the potential patient population for Herceptin increased further recently when it was discovered that the HER2 gene was also implicated in the final stages of prostate cancer in men, which raises the possibility that Herceptin might also be used to treat male cancer sufferers. A treatment for prostate cancer has been to surgically or chemically castrate patients to eliminate the primary source of the androgens that are believed to stimulate the cancer. However, the cancer may eventually return and a study at UCLA recently determined that the cancer comes back because the protein produced by the HER2 gene mimics the male hormones.

The OOPD assert that pharmacogenetic drugs are treated no differently from other drugs. A pharmacogenetic drug that could be demonstrated to be appropriate for use only in a small subset of a larger population, would have no trouble achieving the orphan-drug designation.

Mignon Fogarty, pharmaceutical management consultant at Plan A, Inc. speculates that the orphan drug designation will become less important as pharmacogenetic approaches bring down the cost of drug development as pharmaceutical companies' success rates in drug development are increased. As she points out, the \$500 million cost of bringing one drug to market incorporates development costs for 4,999 other

drugs that fail. The actual cost of developing a single drug is closer to \$75 million. Pharmacogenetics may allow companies to produce more drugs for less money.

Genetic information

The implications of creating genetic pigeonholes for patients have to be addressed. Pharmacogenetics could mean routine screening of generally healthy people seeking medical insurance cover. Legislation might have to be introduced to prevent health insurance providers charging high rates for cover, or even denying it altogether, to people whose genetic profile marks them out as potentially requiring a drug that is more expensive than the one that suits the general population or that makes them likely to suffer severe side effects from a particular drug. In the United States predictive genetic information can be used to deny coverage or to charge a high premium for persons with individual policies. The fear of losing health insurance is a major deterrent to genetic testing.

When the Wellcome Trust carried out Britain's first study on genetic information in life insurance (published in the *British Medical Journal* in 1998) they found inconsistent policies but concluded that any bias based on genetic information was probably due to error or ignorance and not a corporate policy of discrimination. The study looked at the experiences of 7000 members from seven different support groups for families with genetic disorders such as cystic fibrosis and muscular dystrophy. They found that one third had problems getting life insurance, although up to 13 per cent of these cases had no risk on genetic grounds.

Dr Tom Wilkie, the head of biomedical ethics at the Trust, said the study showed that genetic information is liable to be misunderstood. 'There seems to have been unjustified genetic discrimination by insurers in the United Kingdom,' the report stated,

although it was difficult to obtain data on the extent of the discrimination. However, Wilkie noted that the Association of British Insurers had appointed a clinical geneticist as its adviser.

Raymond Woolsey is a researcher at Georgetown University in Washington, DC. It was Woolsey who discovered that the antihistamine Seldane could trigger dangerous heart rhythms. On the subject of genetic information he says, 'It is a medical information issue. You can discriminate against people based on the fact that they have high blood pressure. But people don't think about that as much as they worry about genetic information. We need to communicate to people that genes are just another piece of information.' Ethicist Bartha Maria Knoppers of the University of Montreal recently argued that giving special protection to genetic information makes it more likely to be used in a stigmatising way. 'If we adopt legislation, that only lends credence to the concept that genetic information is different and special,' she said. 'It's in this atmosphere of fear that pharmacogenomics is beginning.'

Iceland

The genetic information of Icelanders is considered to be of particular value by pharmacogenomic researchers because of the extent of inbreeding on the island. Because of its isolated location there may have been little shift in the Icelandic gene pool over the last thousand years. The resulting homogeneity makes disease genes much easier to spot here than in other populations.

On 16 December 1998 the Icelandic parliament passed legislation granting deCODE Genetics of Reykjavik, the right to establish a nationwide database containing health records, genetic and genealogical information of the entire nation created through agreements with hospitals, clinics, and individual physicians to submit their patients' medical records. The company expects its search for disease-causing genes, on which diagnostic tests and therapies could be based, to be greatly enhanced by this

information. The legislation gives deCODE exclusive commercial rights to the data for twelve years, and allows it to enter into agreements with other biotechnology firms or major pharmaceutical companies. Early in 1998, ahead of the legislation being passed, Roche signed a five-year agreement with deCODE to use genetic information uncovered by the research to find new diagnostics and therapies for twelve diseases, including four cardiovascular diseases, four neurological or psychiatric disorders and four metabolic disorders.

The Icelandic government sees genetics as a promising way to generate high-tech jobs for the country's economy. However, the deCODE bill, first introduced in spring 1998, is not without its critics who say it violates basic ethical principles because patients will not be asked for their consent before their records are added to the database. They argue that one company should not have the commercial rights to a whole nation's gene pool. Already, 44 general practitioners and 109 hospital specialists have pledged not to send information to the database unless a patient specifically requests them to do so.

Diagnostics

Within the pharmaceutical industry there are three distinct diagnostic sectors: laboratory diagnostics, representing 70 per cent of the market; point-of-care diagnostics, which covers all testing carried out within a health care system, such as in a doctor's surgery, accounting for 15 per cent of the market, and self-testing, which accounts for the remainder of the market and is largely made up of diabetics' monitoring kits.

Rolf Klasson, president of Bayer Diagnostics' said last year, 'Right now pharmacogenomics is fairly insignificant. In five years it will be important but ten years from now it will be critical.' He predicted that, within a decade, doctors will be prescribing customised drugs, used in tandem with a diagnostic kit to predict a patient's response. 'In the future diagnostic technologies will play an important role hand-in-hand with clinical evaluation. In a health care company there are very strong links

commercially and technologically between a strong diagnostics business and a strong ethical pharmaceuticals business; and they will grow stronger and stronger.’

The first drug to be marketed in parallel with a diagnostic kit was launched in the United States in October 1998. This was Herceptin, touted by Genentech, the biotechnology company that discovered it, as a miracle medicine for breast-cancer sufferers. However, the miracle is only effective for 30 to 40 per cent of patients, specifically those who over express the Her2 protein, which is where the test kit comes in.

In future more medicines are likely to be packaged in the same way as Herceptin. This realisation is prompting pharmaceuticals companies to look again at the diagnostics industry. In 1997 the Swiss company Hoffman LaRoche paid \$11bn to buy the Corange group, the parent company of the world's second-biggest diagnostics business, Boehringer Mannheim. This deal gave LaRoche a 17 per cent market share that made it the world leader in test kits. Sergio Traversa, of the New York-based analysts Mehta Panners, was clear about their reasons for doing so: ‘Pharmacogenomics is where the diagnostics sector is going to expand rapidly... This is the major reason that Roche bought Corange - to have an edge five to 10 years from now in pharmacogenomics.’

In addition to working together on mapping SNPs, Abbott Laboratories, incidentally, the second-biggest diagnostics company, agreed a \$40m deal with French pharmacogenomics firm Genset, in 1997 to develop and market medicines and their relevant diagnostic kits. According to Pascal Brandys, chief executive of Genset, the first pharmacogenomics drugs will be on the market in 2000.

The Human Genome Project

Without the Human Genome Project (HGP) there would be little future for pharmacogenomics. The sequence of the entire human genome is likely to be available in

the near future. The current target date, according to a recent announcement by Dr Francis Collins, director of the National Human Genome Research Institute (NHGRI), is 2002, a year ahead of the revised target of 2003, the 50th anniversary of Watson and Crick's uncovering of the structure of DNA. Already the gene maps are nearly finished, and the project's main goal is to decode the three to four billion base pairs of nucleotides strung along the length of the DNA molecule.

The international consortium involved in the HGP currently includes three laboratories in the United States funded by the NHGRI, which is part of the National Institutes of Health (NIH), the Joint Genome Institute of the United States Department of Energy (DOE), and the Sanger Centre supported in the United Kingdom by the Wellcome Trust.

The full-scale sequencing effort currently being undertaken is based on the success of pilot projects that began a few years ago to test new technologies and strategies for sequencing the human genome. In the pilot phase, eight scientific teams completed the sequencing of over 480 million bases, of which 260 million (close to 10 per cent of the human genome on some estimates) are in high-quality finished form. The sequences have been produced to high degree of accuracy – leading sequencers produce fewer than ten errors for every million bases sequenced.

The HGP had originally planned to complete the human genome sequence by 2005, but the project turned into a race when Celera Genomics of Rockville, Maryland, founded in 1998, announced that it would sequence the genome by the end of 2001. The HGP has promised a 90 per cent complete draft version of the genome by spring 2000. In July 1999 new calculations by Celera appeared to show that the human genome may be as much as one-third larger than previously thought. This new estimate suggests that the number of human genes is likely to be around 100,000, substantially more than earlier estimates of 70,000 to 80,000 and raises questions about whether Celera or the HGP can finish sequencing the genome by their stated target dates.

The 24 September issue of *Science* stated that the team sequencing chromosome 22 were just about to cross the finishing line with an official announcement due in early November. Chromosome 22, with roughly 53 million base pairs, is the second-shortest chromosome. One part of chromosome 22 contains the immunoglobulin-I gene cluster, which is involved in human immune response. Another region includes the Bcr gene, which has been implicated in certain forms of leukaemia.

From its inception in 1990 it has been recognised that the Human Genome Project would raise a number of complex issues for individuals and society as it uncovered more and more information about human genetics. The United States Department of Energy and the NIH have devoted 3 to 5 per cent of their annual HGP budgets to studies of the project's ethical, legal and social implications (ELSI) in an attempt to identify these issues and develop policies that will confront their implications.

Among these are the aspects of the program that are relevant to pharmacogenetics, such as the ability to predict future illnesses well before any symptoms become apparent; the privacy and fair use of genetic information with respect to employers, insurers, banks and others and its possible misuse to discriminate against individuals with a particular genetic make up. The ELSI program recognises that

'One potential outcome of the HGP is that genome research and the wide use of genetic screening could foster a new genetic underclass, leading to a host of new societal conflicts and exacerbating others of long standing.'

*DOE ELSI Program Emphasises Education, Privacy
A Retrospective (1990-1999)*

The fair use of genetic information raises complex issues of access and disclosure. Third parties such as insurers and employers may argue their right to have access to genetic data that can be used to predict a person's susceptibility to illness or their potential need for expensive pharmaceuticals. Many would feel that access to such

information could lead to discrimination against people on the grounds of their genetic make up, a situation that is perhaps not too far removed from discrimination on the grounds of race.

Developments in the science of genomics will greatly accelerate the pace of discovery in pharmacogenetics and should open up a host of possibilities for a wide range of researchers in the fields of pharmacology, physiology, genetics, genomics and medicine. Some analysts predict that within ten years people will be able to carry a record of their entire genome on a small card that doctors will simply slot into an office computers for a quick genetic analysis before prescribing any drugs. Others think that this is rather fanciful and that it is more likely that doctors will test only for specific gene variations that are relevant to the treatment being offered, leaving the rest of the genome unexamined.

DNA chips

Just as important as sequencing the genome, if not more so, is linking each gene to its role in the cell. In 1991 Stephen Fodor and colleagues at Affymax published a paper in *Science* in which they described how they had come up with a scheme to use the same production techniques employed in computer-chip manufacturing to synthesize an array of either short protein fragments or short DNA fragments, called oligonucleotides. The researchers had found a way to generate a large number of compounds quickly, each of which had a unique chemical signature.

DNA arrays are made using a silicon surface coated with linker molecules that bind the four DNA nucleotides. To begin with the linker molecules are covered by a blocking compound that can be removed by exposure to light. The chip is incubated with one of the four bases, binding it to the exposed areas, then the block is reapplied. By repeating this process with different masks and different bases, an array of more than 65,000

different oligonucleotides, each eight base pairs long can be created in just 32 such cycles.

Each oligonucleotide is capable of binding to other fragments of DNA that have complementary sequences. Researchers can isolate the RNA molecules that signal gene expression then label them with a fluorescent tag. The tagged strands are then passed over a DNA array, allowing complementary sequences to bind, while unbound strands are washed away. The strands that bind to locations on the array can be spotted by exciting the fluorescent tags using a laser. As the precise nucleotide sequence of each oligonucleotide in the array is known it is a simple matter to determine the sequence of the gene fragment that has bound there.

DNA chips, or arrays, are two to three centimetre-wide slices of either silicon or glass, upon which there are anything from hundreds to hundreds of thousands of immobilised snippets of DNA. These arrays give researchers the ability to track the expression of many (if not all) of a cell's genes at once, allowing them to witness for the first time the behaviour of thousands of genes acting in concert. One use for these arrays is in tracking cells' responses to drugs. Researchers hope that arrays will help them gauge the success of various drug treatments and tailor medications to a patient's particular genetic make up. The DNA chips will help to pinpoint the genes active in disease, and in metabolising the drugs used to treat it.

Market research surveys would appear to indicate that an annual market for DNA chips in the United States is about \$1 billion and this is likely to grow. In this light it is not surprising that a number of companies have gone into DNA chip production. Molecular biologist Lee Silver, interviewed in the May 1999 issue of *Reason* had this to say about biochips:

This is going to revolutionise medicine. It is also going to revolutionise our understanding of the connection between genes and who people are. For example, say

you are born with a predisposition to a hot temper ... There are probably a multitude of combinations of 30 or 40 different genes that together in certain combinations predispose people to have a hot temper. Using the DNA chip, you can assay 10,000 people to find out the constellation of genes that predisposes you to a hot temper, or predisposes you to being depressed, or predisposes you to any kind of trait. That is phenomenal. Most people right now don't understand the power of this yet ... We are waiting for the Human Genome Project to finish; then we can take those 70,000 genes and put them on a DNA chip.

Unstated in the article is quite what would be done with this information but given the current vogue for a chemical solution to behavioural problems, and a culture in which 'happiness' is seen as a default state, it is not difficult to imagine a pharmacogenetics research lab somewhere searching for a 'cure' for bad temper.

Researchers at the University of Chicago's Argonne National Laboratories. have been working with a team of scientists from the Russian Academy of Sciences' Engelhardt Institute of Molecular Biology to develop ways to produce a new super-efficient biochip that can sequence DNA thousands of times faster than current methods. Several companies are now competing to produce biochips. Motorola Inc.'s Bio-Chip Systems Unit is building biochips and their associated systems and should be producing prototypes next year. The unit chief is Nicholas Naclerio, formerly of the Defence Advanced Research Projects Agency. The company has recruited life scientists and licensed technology from Argonne National Laboratories and the Engelhardt Institute.

Motorola are focusing on bioarrays and microfluidic chips. Numerous tiny probes on the bioarrays store molecules such as DNA and can carry out several different tests at once to identify, for example, genetic markers that correspond to responses to a new drug. Naclerio compares microfluidic chips to microprocessors as they can be programmed to carry out complex chemical processes, a 'lab-on-a-chip' he calls it.

Earlier this year Xenometrix granted a worldwide license for gene expression profiling to Motorola that covers the collection of gene expression profiles utilising all platforms, including microarrays.

One reason that biochips have been slow to come to the market place may be because most are made of silicon, which is expensive to manufacture and toxic to many biochemical processes. Marc Madou, professor of chemistry and materials science and engineering at Ohio State University and chief scientist for Microbionics Inc. in Menlo Park has created a disposable biosensor chip from plastics and polymers that can be mass produced on continuous large plastic sheets. It is hoped that these chips will cost one-tenth the amount of existing silicon-based chips. Madou's biosensor could be placed in a hand-held device and connected to an online computer in the doctor's surgery and be used to analyse a drop of blood. Each biosensor chip would cost less than a dollar and would simply be thrown away and replaced after use.

Genetic polymorphism

Traits such as ABO blood groupings are examples of genetic polymorphism - that is a relatively common monogenic characteristic that is expressed in the general human population in at least two phenotypes, each of which occurs with a frequency of greater than 1 to 2 per cent. The ability to identify the genetic variations between people that influence their differing responses to drugs is at the heart of pharmacogenetics. In the context of drug development genetic polymorphism of drug metabolising enzymes gives rise to a diversity of responses to drugs among the population with two or more distinct subgroups with differences in their ability to metabolise particular drugs.

Drug metabolism

Functionally significant variations have now been identified for many of the enzymes involved in drug metabolism, as well as for target receptors and enzymes that are involved in drug responses. A great deal of pharmacogenetics research has focused on the mechanisms that determine systemic drug concentration: the end result of ingestion, absorption, metabolism, clearance and excretion of a drug. But drugs also act on receptors that can themselves have polymorphisms. Pharmacogenetics researchers have to identify all of the proteins that medicines encounter in the body and determine how these proteins vary from person to person. This means identifying and scrutinising the genes that code for these proteins to establish the basis for the protein differences.

The primary source of metabolic variation between individuals appears to be the polymorphic expression of several drug-metabolizing hepatic enzymes. The activity of drug-metabolizing enzymes plays a major role in both the intensity and the duration of drug action. Drugs are altered in the liver to increase the rate at which they are eliminated from the body, and metabolism is frequently the decisive factor in determining when the drug will be removed from the body.

There are many drug-metabolizing enzymes that can be shown to have polymorphic expression, but the enzymes that are mainly involved in hereditary variations in drug metabolism belong to the cytochrome P450 superfamily. Perhaps the most important of the polymorphically expressed P450 enzymes is CYP2D6, which is responsible for the metabolism of many of the older tricyclic antidepressants, as well as many of the newer antidepressants, such as the selective serotonin reuptake inhibitors. These are some of the most frequently prescribed drugs on the market, yet polymorphic variability in the population makes it difficult to predict the response to a given dose. Certain alleles of the CYP2D6 gene are the cause of slow metabolism of the drugs and have been found to be responsible for some of the characteristic side effects of these drugs.

The human population can be divided into two groups according to the polymorphic expression of at least three P450 enzymes: 'poor' metabolisers (PMs) or PM-phenotypes, who express dysfunctional or inactive enzymes, and 'extensive' metabolisers (EMs) or EM-phenotypes, who express enzymes that show normal activity. The ultra-rapid metabolisers (UM-phenotype), a much rarer group, have an increased capability for drug metabolism. PMs account for 3 to 7 per cent of whites. The frequency of these polymorphisms can be very different in other ethnic groups (a subject that is the province of pharmac anthropology). The rate at which many drugs are metabolised may vary 10- to 100-fold between PMs and EMs.

EMs have at least one wild-type allele and make a sufficient quantity of functioning metabolic enzyme to support normal drug metabolism. PMs have no wild-type allele and do not produce an active gene product. It is differences in metabolic rates between individuals that alter the expected relationship between the dose of a drug and its concentration in the blood or the length of time it stays in the blood. This is the reason why administering a 'typical' dose of a drug to some individuals can result in a prolonged therapeutic effect or drug-related toxicity as polymorphic differences in metabolising enzymes leads to a failure to clear a drug from the blood or alters the pattern of metabolism with the result that toxic metabolites are produced.

Polymorphisms in PMs, therefore, result in a genetic predisposition to adverse drug effects.

A good example is the variability observed in the metabolism of azathioprine, a treatment for leukaemia and autoimmune disorders. The major metabolic route of azathioprine involves the enzyme TPMT. About 10 per cent of Caucasians carry an allele of the TPMT gene that encodes an inactive protein. As long as these people are heterozygous for this trait and also carry a normal allele they produce the normal enzyme and are able to metabolise azathioprine. However, less than 1 per cent of

people, have an inactive allele on both chromosome sets and so cannot metabolise azathioprine efficiently. When such individuals are treated with the drug levels of azathioprine in the blood become very high, and this leads to acute bone marrow failure. A genetic test is available to determine whether a patient is homozygous for the inactive allele. Having thus pinpointed those patients with the TPMT deficiency, an alternative therapy may be sought or the azathioprine dose administered may be reduced. This pharmacogenetic test has clear value to potential azathioprine patients. The number of patients who can benefit from such a test remains low, however.

Now that extensive mapping and analysis of the genetic code is under way, researchers can begin to identify genes that might influence the effectiveness of a drug. The first pharmacogenetic attribute to be identified was phenylthiourea 'taste blindness'. This was the first indication that individuals could be identified by variations in their sensitivity to chemicals and that this attribute was heritable. Black Africans had an incidence of taste blindness of around six per cent, but Black Americans ranged from two to twenty-three per cent, white Americans, around thirty per cent, Chinese around six per cent and Eastern Eskimos around forty per cent.

Alcohol sensitivity is another trait that has been recognised for a long time. One of the first observed examples of genetic polymorphism in drug metabolism was the difference in alcohol tolerance between Caucasians and Asian populations. Genetic variations in the production of ALDH (aldehyde dehydrogenase) and ADH (alcohol dehydrogenase), enzymes involved in the metabolism of alcohol, are very common in Asians, particularly among the Japanese.

It can be shown that the frequencies of many pharmacogenetics traits within a population are linked to ethnicity. Knowledge of population variations in pharmacogenetic characteristics is essential for new drug development and clinical care.

Determining polymorphism

Genetic polymorphism is determined by two methods: molecular genotyping and phenotyping. Molecular genotyping identifies the genetic make up of an individual using a variety of DNA based methods. Phenotyping involves the administration of a probe drug to an individual followed by analysis of specific metabolites in blood or urine monitored over time.

Molecular genotyping can be used to screen for genetic predispositions to disease. The advantage of this type of screening is that it only has to be determined once in the lifetime of the patient. Molecular genotyping is also used to identify poor metabolisers prior to beginning drug therapy. Doctors can screen their patients for the PM genotype before administering drugs that are known to have little therapeutic benefit or even to cause unacceptable adverse reactions in poor metabolisers. This is especially true for treatment with cardiovascular and antipsychotic drugs where polymorphism has been shown to have significant clinical consequences. In the near future pharmacogenetic profiles may be routinely used in determining the type of drug to be prescribed and its dosage before therapy is undertaken.

In 1998, the National Human Genome Research Institute in the United States coordinated the assembly of a DNA Polymorphism Discovery Resource of anonymous samples from 450 United States residents with ancestry from all the major population groups of the world. The purpose of the resource is to aid in the discovery of DNA sequence variation. It was set up solely for the purpose of determining human variation and no medical, phenotypic, or demographic information was linked to individual samples. The resource is publicly available and stored at the National Institute of General Medical Sciences Human Genetic Mutant Cell Repository at the Coriell Institute for Medical Research.

The Pharmacogenetic Research Network

In 1999 the National Institute of General Medical Sciences sponsored a Request for Applications for a 'Pharmacogenetic Research Network and Database'. The object of this is to solicit applications to study functional variation in genes and proteins that play essential roles in individual drug responses and to make predictions about phenotypic responses based on genotypic make-up. The Pharmacogenetic database will be designed to accommodate the information gathered about a particular gene and the protein it encodes, linking sequence variations in the gene to biochemical changes in the encoded protein and the consequences for drug response. The database is intended to be used as a research tool by a broad range of scientists. Research groups are specifically encouraged to make use of the NHGRI DNA Polymorphism Discovery Resource.

Many pharmaceutical companies have drugs in development that are likely to show variations in effect due to polymorphic metabolism. Antidepressants, antipsychotics and cardiovascular drugs all come within this category. Increasingly, clinical trial subjects are being screened for specific genetic mutations to ensure that the study population is both relevant and representative for the drug under test.

Pharmacogenetics and the FDA

In April 1997 the Center for Drug Evaluation and Research (CDER), a branch of the United States Food and Drug Administration (FDA), issued its Guidance for Industry entitled 'Drug Metabolism/Drug Interaction Studies in the Drug Development Process: Studies In Vitro'. The FDA supports pharmacogenetic testing throughout drug development, recognising that 'Pharmacogenetics already has influenced therapeutics.' When a genetic polymorphism affects an important metabolic route of elimination, dosing adjustments may be required to achieve the safe and effective use of a drug. The FDA believes that 'Identifying metabolic differences in patient groups based on genetic polymorphisms ...will provide improved dosing recommendations in product labelling

... allowing prescribers to anticipate necessary dose adjustments. Indeed, in some cases, understanding how to adjust doses to avoid toxicity may allow the marketing of a drug that would have an unacceptable level of toxicity were its toxicity unpredictable and unpreventable.'

It is the opinion of the FDA that 'it is difficult to justify marketing a drug without knowing how it is metabolised' and therefore, 'When a genetic polymorphism affects an important metabolic route of elimination, large dosing adjustments may be necessary to achieve the safe and effective use of the drug.' With this in mind, the FDA recommends that appropriate metabolic studies, including in vitro and pharmacogenetics testing, be undertaken throughout the drug development process.

Single nucleotide polymorphisms

The collection of DNA that makes up the human genome is made up of about three billion base pairs of nucleotides. A mutation that is capable of conferring a risk for disease, or a changed metabolism for a drug, can be as small as a single altered base pair – a single nucleotide polymorphism, or SNP (pronounced 'snip'). Recently Aravinda Chakravarti of the Case Western Reserve University School of Medicine in Ohio announced that his team had found 874 different SNPs that might be associated with high blood pressure. Of these, more than half resulted in a change in the protein controlled by the gene.

For reasons yet to be uncovered, certain genes contain much more natural variation, which means more SNPs, than others. There is no certainty that any or all of the SNPs in a particular gene, are indicators of disease susceptibility or variations in drug response, but the hope is that SNPs will prove to be a powerful tool for tracing disease genes and developing individualised drugs. SNPs are believed to be evenly distributed

along the human genome at a frequency of about one every 300 to a thousand bases. Once found, the SNPs will be mapped to positions on the genome.

Particular alleles tend to be accompanied by particular patterns of SNPs and that information can perhaps be correlated with any diseases, or side-effects of treatments, that the individual may suffer from and, given a reasonable sample of sufferers and non-sufferers, the genes associated with particular diseases and side-effects can be tracked down and their various alleles sequenced. Pharmacogenomic studies should be able to show which combinations of alleles lead to polygenic diseases.

Genset, in France, is developing a map of the human genome by testing the DNA of more than 100 people. Dr Daniel Cohen, Genset's chief genomics officer, devised the first rough physical map of the human genome in 1993. The Genset map will contain 60,000 SNPs that are within or near genes associated with disease or differing drug reactions. Genset are working with US pharmaceutical firm Abbott Laboratories, which has invested \$20 million in the French company. Abbott and Genset plan to market their SNP data to drug companies that need to locate variant nucleotides shared by subjects who do not respond to a drug during clinical trials. The information so gained could then be used to formulate tests that would allow unresponsive patients to be removed from the trials. As yet, only a few pharmaceutical companies are currently screening individuals for specific polymorphisms before entering them into clinical trials to ensure that the study population is representative of the general population.

Genset's genome map enables them to compare DNA sequences between individuals who differ in a particular characteristic, such as a specific drug response. Comparing gene sequences of a group of responders to a group of nonresponders allows researchers to identify the gene and which allele of that gene is responsible for the characteristic being studied. This is pharmacogenomics rather than pharmacogenetics as the genes can be found without any knowledge of the biochemistry of the response.

This technology can be applied to a wide variety of drug responses: adverse side effects such as heart valve abnormalities resulting from diet drugs, or cholinergic effects of tricyclic antidepressants or the failure of selective serotonin reuptake inhibitor antidepressants or interferon-alpha for hepatitis C infection might all have significant genetic ingredients that can be found through gene comparison studies.

The Wellcome Trust, in partnership with ten major pharmaceutical firms in the United States and Europe, Bayer Group AG, Bristol-Myers Squibb Co., Glaxo Wellcome PLC, Hoechst Marion Roussel AG, Monsanto Co., Novartis AG, Pfizer Inc., Roche Holding Ltd., SmithKline Beecham PLC, and Zeneca Group PLC, has set up The SNP Consortium (TSC). The consortium plans to create a SNP map of the whole genome over the next two years at a cost of \$45 million.

The goal is to have 150,000 SNPs mapped in this way by mid-2001. TSC researchers hope that compiling a database of several hundred thousand SNPs will make it easier to track smaller segments of the genome and identify patterns of inheritance that affect health. Few SNPs are likely to be directly involved in disease, but the SNP map may make it possible to diagnose illnesses earlier and avoid giving drugs to patients likely to experience side effects.

The work of compiling the map is being carried out by three genome sequencing teams: the Whitehead Institute for Biomedical Research at the Massachusetts Institute of Technology in Cambridge, Massachusetts, headed by Eric Lander, the Sanger Centre near Cambridge, UK, under David Bentley, and the Washington University Genome Center in St. Louis under Elaine Mardis. Discovered SNPs are sent to the Cold Spring Harbor Laboratory in New York where the data is double-checked and computer cross-matched against already mapped human genome sequence data. Wherever a match is found the genomic location can be mapped.

The data will be made available to the public with no preferential access for consortium members. Every quarter (the first release was on 15 July 1999) the Cold Spring Harbor Laboratory will post the data on a public Web site.

Researchers in Japan are about to embark on an SNPs project of their own that will involve using samples from 50 Japanese people to map between 100,000 and 150,000 SNPs, concentrating on areas associated with gene expression and function at a cost of \$50 million over 2 years. Researchers hope to increase the likelihood of spotting SNPs associated with disease susceptibility and drug response. The research will target diseases prevalent in Japan, including cancer, diabetes, rheumatoid arthritis and cardiovascular diseases. Drug companies hope that studies of SNPs will reveal why medication that proves effective in one person is useless or produces side effects in another.

The teams headed by Aravinda Chakravarti at Case Western, and by Eric Lander, at the Whitehead Institute, are taking a different approach. Rather than attempting a whole-genome map they are gathering SNPs from a set of some 200 genes related to hypertension and other multigene diseases from more than 125 individuals. Both teams found that SNPs within the coding region of genes that alter the composition of the encoded protein are very rare in the general population. Chakravarti reckons that, 'There seems to be a strong selection against any change in protein structure. [Most of these changes] have been weeded out in the course of evolution.'

Chakravarti and colleagues are now working with people who have high blood pressure to link the SNPs with actual disease. His team has estimated the total number of SNPs in human genes as being close to a million. However, Lander's team believe that there are 240,000 to 400,000.

Lander's study reports that about 10 per cent of the protein-altering SNPs seem to be specific for certain subpopulations, such as Asians or African-Americans.

They looked at how SNPs in 106 genes suspected of involvement in coronary artery disease, type II diabetes and schizophrenia differ in people of European, African-American, African Pygmy and Asian descent. Most differences in SNPs have nothing to do with such groups, although there are a small number of variations that do seem to be linked with population group.

Pharmacogenetics and environmental medicine

As has been mentioned throughout this paper, susceptibility to diseases and the efficacy of their treatments are consequences of both genetic and environmental factors. Identifying the genetic variants that influence an individual's response to toxins in the environment can allow doctors to identify those who are at greatest risk from exposure. The environmental agent could be anything from an industrially produced chemical with carcinogenic properties to a naturally occurring virus.

Environmental pharmacogenetics can become much more complicated than clinical, drug-response pharmacogenetics. Carcinogenesis usually involves the action of many different genes and the identity and exposure levels of carcinogens in the environment may well be unknown, in contrast to a well-defined and readily identifiable viral or bacterial disease agent. Behavioural considerations would probably also have to be taken into account in assessing the significance of a particular polymorphism regarding susceptibility to cancer.

Most environmental carcinogens are metabolically activated or inactivated by variants of cytochrome P450 enzymes. Animals in which these enzymes have been inhibited in laboratory studies have shown an increase in cancer induction. Some human population studies have also shown that cytochrome polymorphisms are linked to a higher incidence of various cancers. One enzyme has several known polymorphisms that have

been linked to cancers of the lung, stomach, liver and nasopharynx, although the results of numerous trials have shown conflicting results.

The possibility exists that people could be routinely screened and given an 'environmental risk factor' rating. There are obvious consequences of such a development for employment. People applying for jobs within the chemical industry, for example, would doubtless be rejected if their risk factor exceeded some arbitrarily set limit. The question would then arise as to whose risk it was to take and it is easy to envisage people desperate for employment of any sort evading or refusing to be tested. Issues of confidentiality are involved where, for instance, a person has been genetically screened prior to treatment for a non-related medical condition. Would the results of the screening be made available to the employer? Could the employer refuse to consider an application simply on the grounds that they knew screening had been carried out but they had been refused permission to view the results? A case of guilty until proven innocent for the prospective employee?

Many employers already undertake routine pre-employment health assessments as part of the employee recruitment procedure. A Health and Safety Executive research study on health surveillance based on a sample of over 1,600 employers reported that just under one third of employers reported that they were carrying out such assessments of prospective employees. Given the limited predictive value of genetic screening at present and the fact that most people spend a relatively short time in a particular job these days it does not seem particularly reasonable to use these tests as the basis for selecting an employee.

In 1993 the Nuffield Council on Bioethics concluded that:

genetic screening of employees for increased occupational risks ought only to be contemplated where:-

- i) there is strong evidence of a clear connection between the working environment and the development of the condition for which genetic testing can be conducted;*
- ii) the condition in question is one which seriously endangers the health of the employee or is one in which an affected employee is likely to present a serious danger to third parties;*
- iii) the condition is one for which the dangers cannot be eliminated or significantly reduced by reasonable measures taken by the employer to modify or respond to the environmental risks.*

A survey carried out recently by the disability charity Radar found that some 90 per cent of disabled people questioned wanted a ban on employers and insurers being given access to information suggesting someone might develop a genetic condition. Radar found people feared the growing emphasis on genetics would lead to further discrimination.

At present the only employer currently carrying out routine genetic screening is the Ministry of Defence. All applicants for air crew training are screened for sickle cell disease and trait. This is designed to protect the individual and others from the potentially dangerous effects of a sickling crisis provoked by low oxygen pressures in flight. It is believed, although the evidence is contested, that individuals with sickle cell trait can develop symptoms of sickling if exposed to very low oxygen pressures.

Alzheimer's disease

It is estimated that four million Americans now suffer with Alzheimer's disease and by 2040 the number has been predicted to rise to nine million if no cure or treatment is found. It is the fourth leading cause of death in the United States.

Alzheimer's disease has been subjected to intense genetic analysis. Genetically speaking it is a heterogeneous disorder associated with three determinative or causal

genes and one susceptibility gene. The three determinative Alzheimer's genes are dominant – their presence in the genome is a guarantee that the disease will occur, usually in the early-onset form between the early forties and mid-fifties. Less than 1 per cent of all cases take this form.

There is no predictive genetic test for the more typical late-onset form of Alzheimer's. In 1993 the apolipoprotein gene was linked to 50 to 60 per cent of cases of late-onset Alzheimer's. Apolipoprotein E (ApoE) is involved in the transport of cholesterol and phospholipids and is implicated in synaptic remodelling and regeneration. Some researchers see an association between having the E4 form of the gene and developing the disease. However, the presence of the allele is not useful as a risk test except perhaps for the 2 per cent of people with two of the alleles in question, and even then there is no certainty of eventual illness.

Robert Butler, editor of *Geriatrics*, urged clinicians to be cautious, emphasising that genetic testing has not yet been established as a reliable predictor and that discrimination in employment and insurance was likely if screening of this nature became widespread.

Judes Poirier, Director of the McGill Aging Research Center at McGill University does believe that pharmacogenetics can play a role in the treatment of Alzheimer's – genetic information can be used to prescribe the right drug to the right patient. He says that a natural question to ask is whether a person's ApoE genotype would affect his or her response to memory-enhancing drugs. A study carried out by Poirier suggested that if a placebo is given to two groups of subjects, one being homozygous for E4 and the other lacking E4 altogether, a statistically significant difference will be seen in the two groups simply because Alzheimer's is a disease with two distinct rates of degradation. Those patients lacking E4 did not develop Alzheimer's until age 85 but their rate of degradation was around two to three times faster.

Poirier found that non-ApoE4 subjects responded quite well to Tacrine, while the ApoE4 subjects did not. Tacrine works by blocking the enzyme that degrades acetylcholine. Another drug tested, Xanomeline, which replaces acetylcholine, would be expected to work for everyone. Unexpectedly, Poirier found that patients with two copies of ApoE4 actually did worse than those taking a placebo. Drugs designed to stimulate the cholinergic system tend to work well in the non-E4 patient, whereas those agents that are non-cholinergic will work in the E4 subject.

What of the future?

Pharmacogenomics may not after all be the bright future of medicine. William A. Haseltine, chairman and chief executive of Human Genome Sciences in Rockville, Maryland, is unconvinced for one. He contends that the multiple genes involved in a drug reaction can be hard to decipher and that diagnostic tests can be unreliable. Some patients could still suffer life-threatening reactions to medication. In a debate held at Bio 98 reported in *Nature Biotechnology*, October 1998, he said:

'Anybody who has looked at the reliability of diagnostic tests knows they're lousy. Are you then going to not treat somebody effectively because your diagnostic test was wrong or, even worse, are you going to treat somebody with a drug that may kill them? Are you going to accept the liability of having a diagnostic test to match your drug that targets in on a small profile? I don't think so.'

In 1993 Harvard Emeritus Professor of Biology Ruth Hubbard and Elijah Wald wrote an influential book called *Exploding the Gene Myth*. They examined possible negative consequences of genetic diagnoses, including people being refused employment or insurance on the basis of their genetic profile. A basic problem, Hubbard and Wald argue, is that genetic tests and modifications encourage us to look upon ourselves as a

collection of tiny genetic parts, rather than as whole human beings who are affected by their environment as well as their genes.

Environmental factors, such as diet, other drugs that may be prescribed at the same time, gender and overall state of health, have to be considered along with the genome when assessing the way in which a patient responds to a drug. 'You've got to consider the whole person when using a drug. The pharmacogenomic argument is very similar to the sociobiology argument that everything is in the genes, when it is not,' said Haseltine.

Haseltine also fears that the focus may shift towards pinpointing the people who are 'genetically right' for the drugs pharmaceuticals companies want to sell and away from finding the right drugs to treat individuals. 'That's not where we want to go,' he says. 'We still want new drugs that treat as many people as possible.'

Terfenadine, antibiotics and grapefruit juice cause heart arrhythmia if taken concurrently. It is conceivable that the effect shows up only in a genetically defined subset of the population, but it appears implausible at least at the present time, that pharmacogenetics could have identified this problem in advance.

The challenge of pharmacogenetics

The Human Genome Project along with a number of rival commercial and academic efforts such as that of Celera Genomics looks set to provide a comprehensive map and sequence of the human genome within the next two to three years. The development of techniques in genetic engineering has given us the ability to identify and isolate many genes that are important to the understanding of disease. The pharmaceutical industry is being flooded with information as it enters a period both of prodigious complexity and unprecedented opportunity. The challenge for the industry will be in selecting the best therapeutic targets among the huge number that present themselves. The future of

drug development may well lie in the efficient identification of these targets that are most receptive to therapeutic attack.

What difference can we expect to see to our health care provisions in the light of developments in pharmacogenetics and pharmacogenomics? Within the next five to fifteen years the identification of genetic polymorphisms that are relative to the diagnosis and treatment of disease will bring about more efficient clinical trial design. Drugs may reach the market faster as a result, and stay there longer. Pharmaceutical companies might look again at their past failures. A subpopulation able to be treated safely on account of their particular genetic make up might be found for drugs that had resulted in too many adverse reactions in previous clinical trials. As in the case of Herceptin, more and more drugs will be marketed alongside diagnostic tests to identify those individuals whose genotype best suits them to the treatment being offered.

Although there will undoubtedly be a number of highly effective drugs marketed exclusively to small population groups further into the future the focus may shift as pharmacogenomics is used to identify sections of the population with minimal variation. After all it is hard to imagine the pharmaceutical industry continuing to market drug therapies for increasingly smaller patient populations, with or without orphan drug legislation. The knowledge gained by pharmacogenomics could be used to develop extremely effective, broadly tolerated drugs that can be prescribed to the largest possible population.

There are obviously questions that will need to be addressed and discussed in as wide a range of forums as possible, including the scientific and medical communities, health care practitioners and patient support groups as well as in society as a whole if the new technology and knowledge are to be used in the most effective and equitable way possible to provide the best environment for safe, cost-effective, and effective medical treatment. Especially these questions will need to be directed towards issues of confidentiality regarding the gathering of genetic material and the avoidance of

discriminatory practises in the utilisation of that material. Some assert that the fully decoded human genome may prove that everybody is predisposed to some kind of abnormality. If this were the case then would it eliminate any logical basis for discrimination?

The New Jersey Genetic Privacy Act, passed in 1996, could be the first of many state and national laws in this field. It prevents employers and insurance companies from discriminating against people on the basis of genetic tests. In addition the law also makes genetic information the personal property of the individual and requires informed consent before it can be used. If a person requests it, in most cases DNA samples are to be destroyed promptly.

The New Jersey law has yet to be put to the test in the courts. In fact, few if any genetic discrimination cases have been brought. As Deborah Lockner-Doyle, president of the National Society for Genetic Counselors in Seattle acknowledges, 'The magnitude of the problem is quite small, but the fear of the problem is quite great.'

It had been expected, for example, that many women would want to be tested for defects in such genes as BRCA I and BRCA 2 that can lead to breast and ovarian cancer, but the demand has not been as large as was anticipated. As Barry M. Berger, Director of Pathology and Laboratory Medicine at Harvard Vanguard Medical Associates put it, 'There is probably nothing as intensely private as your genotype and people fear that this information will get misused.'

The promise of pharmacogenetics is as yet unfulfilled but it is one that cannot be ignored. The impact of genetic information on the future of drug development will be profound.

Background Reading List

Books

Hubbard, R. and Wald, E., *Exploding the Gene Myth*, Beacon Press, 1993.

Katz Rothman, B., *Genetic Maps and Human Imaginations: The limits of science in understanding who we are*, W.W. Norton and Company Ltd., 1998

Kalow, W., *Pharmacogenetics of Drug Metabolism*, Pergamon, 1992.

Marks, J., *Human Biodiversity: Genes, Race and History* (in the series: Foundations of Human Behaviour), Aldine de Gruyter, 1995.

Weber, W. W., *Pharmacogenetics*, Oxford University Press, 1997.

Wilkie, T., *Perilous Knowledge: The Human Genome Project and Its Implications*, Faber and Faber Ltd., 1993.

Articles

Peter L. Bullock, "Pharmacogenetics and Its Impact on Drug Development", *Drug Benefit Trends* - available from Medscape

James Kling, "Opportunities Abound in Pharmacogenomics", *The Scientist*, Volume 13, no.10, 10 May 1999

National Institute of General Medical Sciences, "Medicines by Design: The Biological Revolution in Pharmacology – an overview of drugs of the future" NIH, Bethesda, Maryland 20892-6200

Brian B. Spear, "Pharmacogenomics: Today, Tomorrow, and Beyond", *Drug Benefit Trends* - available from Medscape

Garry Stix, "Personal Pills", *Scientific American*, October 1998

Janice Hopkins Tanne, "The New Word in Designer Drugs", *British Medical Journal* 27 June 1998

Websites

“Healing the Helix: The Future of Genomic Medicine”, from Biomednet's HMS Beagle, <http://www.biomednet.com/hmsbeagle/45/resnews/meeting.htm>

“Professionals' Resource Guide to Genetics Education for Primary Care Providers and Other Health Care Professionals”
<http://www.cc.emory.edu/PEDIATRICS/corn/corn.htm>

The Gene Letter , an electronic newsletter about genetics, ethics and public policy,
<http://www.geneletter.org/index.htm>

“Medicine and the New Genetics” , Human Genome Project, contains many excellent links, <http://www.ornl.gov/hgmis/resource/medicine.html>

“Toward the 21st Century: Incorporating Genetics into Primary Health Care”,
<http://www.ornl.gov/hgmis/publicat/hgn/v9n1/15cshl.htm>

“Genetic Secrets: Protecting Privacy and Confidentiality in the Genetic Era”,
<http://www.ornl.gov/hgmis/publicat/hgn/v9n1/16secret.html>

Human Genome News --the newsletter of the Human Genome Project,
<http://www.ornl.gov/hgmis/publicat/publications.html#hgn>

“The Implications of Individualizing Medicine Through Genomics” a white paper from the Stanford University Program on Genomics, Ethics, and Society,
<http://www.stanford.edu/dept/scbe/individu.htm>

“Medical Genetics, applications of genetics to clinical practice”,
<http://medgen.genetics.utah.edu>

Issues and questions arising from the background paper

Social, ethical and public policy research themes

This section identifies issues and questions arising from discussions in this paper. It is neither exhaustive nor complete, but rather is meant to stimulate discussion.

- What are the wider social implications of creating genetic pigeonholes for patients? What is the impact of genetic screening on the way society views and supports people with identified susceptibilities, chronic illness or disabilities?
- A consequence of pharmacogenetics may be a greater emphasis on therapies which rely on genetic profiling. Are social measures to ameliorate disease being short-changed? What are these social factors and social measures?
- What is the social impact of the routine screening of generally healthy people with the expanded use of predictive genetic information?
- Will more sophisticated genetic information lessen or increase the basis for discrimination? What social mechanisms would tip the balance one way or the other?
- What happens when there is a test available to identify individuals who would/would not benefit from a particular drug treatment but the test would prove valuable for only a small percentage of the population?
- As environmental medicine develops further, will pre-employment screening become more attractive for employees/employers? If so, how is this happening? Will workplace issues of safety themselves influence the direction of pharmacogenetics research and development?
- Knowledge of population variations in genetic or biochemical characteristics is essential for new drug development and clinical care. Information is linked to population groups identified by ethnic categories. What are the social and scientific assumptions of using categories which refer to culturally defined ethnic groups, and are they problematic? What happens in the use of these categories? This issue is conceptually complicated, and it is necessary to distinguish both in genetics and in sociology between populations and individuals. Is there a risk that predictive

information derived from populations (which might legitimately be applied to an individual in a medical emergency) might be unthinkingly and illegitimately applied to individuals as a matter of routine?

- Smaller concerns are being formed specifically to take advantage of the legislation on orphan drugs and to develop products for a tightly focused market. Coupled with pharmacogenetics will developments change the objectives of the pharmaceutical industry? If so, how?
- Can pharmacogenetics simultaneously deliver cost savings to health care providers and a satisfactory return on investment to shareholders in pharmaceutical companies?
- Pharmacogenomics may in the long run provide less costly treatments and improvements in the identification of an effective therapy. How will these fit into existing health care delivery systems? Will private health care organisations see advantages over their competitors, if they are the first to offer tailored health care to customers?
- What effect will increased knowledge about differences in individual genetic profiles have on health service provision (for example, when a minority of individuals require a drug that is more expensive than one that suits the general population)?
- Does attention to personal responsibility for health become greater with routine screening? Does it diminish collective NHS provision?
- How is the insurance industry responding, if at all, to these developments? What effect is it having on individuals and groups seeking life coverage or private medical coverage in the UK?
- The US Food and Drug Administration has recommended that pharmacogenetics research and testing be integrated into the drug development process. What is the regulatory response in the UK and at the level of the EU?
- In the US, the state of New Jersey has passed one of the earliest laws prohibiting discrimination in employment and insurance on the basis of genetic tests. The law makes genetic information the personal property of the individual, and requires

informed consent for it to be used. Samples are, in most cases, to be destroyed promptly after immediate use. Is giving genetic information a special status in law desirable? Can (and should) a coherent law on the uses of genetic information be formulated for the UK? How would this affect the establishment of collections of human DNA and tissue banks required for research purposes?

- Selective serotonin reuptake inhibitors (new antidepressants) are some of the most frequently prescribed drugs on the market, and they are of interest to pharmacogenetics researchers. As more sophisticated diagnosis of drugs become available, what will be the short term and long term implications on (i) mental health treatment (ii) access to drugs (iii) pharmaceutical therapy bias in mental health care (iv) the effect on psychosocial interventions, if any?