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The role of molecular biology in pharmacodynamic research

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Abstract

Molecular biology has the potential to influence virtually every aspect of the discovery, development and use of drugs. Gene expression analysis, 'reverse' pharmacology and the use of genetic polymorphisms provide important new approaches to identifying drug targets. Unravelling the human genome has the potential to reveal the genetic basis of complex disorders, and in so doing, improve disease risk stratification and the way in which we individualise drug therapy.

Keywords: Drug target; Polymorphism; Hypertension; Therapeutics; Pharmacogenetics

1. Introduction

In this review, the intention is much more to introduce the reader to the role of molecular biology in relation to pharmacodynamics, than to provide a comprehensive account of this subject. We have chosen to illustrate the topic through examples drawn from cardiovascular research, especially in hypertension.

2. Finding new drug targets

2.1. Gene expression analysis

Knowledge of the pattern of expression of genomic information is essential to the understanding of biological processes [1]. Patterns of gene expression in the tissues of

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patients will differ from those in the normal tissues of controls. Analysis of these differences provides a powerful tool for elucidating drug targets. Although effective drugs have been developed against protein targets that are widely expressed in the body, aiming for targets that are expressed only in selected tissues may limit the potential for unwanted effects. It is important to remember that, ultimately, it is proteins that effect cellular functions, not genes. The study of the human 'proteome' will be much more complex than that of the human genome but will complement gene expression studies and is beginning to be addressed by the emerging field of proteomics [2].

There are many ways in which gene expression can be investigated. For example, so-called expressed sequence tags (ESTs) provide a complementary DNA (cDNA) representation of cellular messenger RNA (mRNA) from a cell or tissue of interest [3–5]. A cardiovascular EST resource has been developed using adult and fetal heart, hypertrophic heart, ischaemic heart, and adult aorta [6]. Comparison of the EST profiles of hypertrophied heart and normal heart has revealed up to 90 individual genes that may be down-regulated in hypertrophied cardiac tissue. The largest decreases are observed in transcripts encoding cell-structure proteins, such as those of the collagen family. Two hundred genes that might be up-regulated were also identified.

Serial analysis of gene expression (SAGE) is essentially an accelerated version of EST sequencing [7,8] and has been used to investigate gene expression in endothelial cells in response to an atherogenic stimulus [9]. In this in vitro study, endothelial cells were activated with a medium of human monocytes that had been stimulated with moderately oxidised low-density lipoprotein. This supernatant also contained numerous cytokines, chemoattractants and growth factors, resembling the early endothelium atherogenic stimulus. SAGE analysis detected 56 genes that underwent altered expression, of which, 42 were known, such as interleukin-8, vascular cell adhesion molecule 1 and E-selectin.

cDNA microarrays are also used to analyse genome-wide patterns of mRNA expression [10], as well as to establish genotypes and detect polymorphisms [11]. This technology has been applied to the investigation of gene expression in the vascular smooth muscle cells, which may have an important role in conditions that alter arterial mechanics, such as hypertensive vascular disease. A DNA microarray containing 5000 genes with putative functions in blood pressure control was used to investigate the transcriptional profile of human aortic smooth muscle cells undergoing mechanical deformation [12]. In addition to vascular endothelial growth factor, which was used as a control, only three transcripts were induced more than 2.5-fold. These were cyclooxygenase-1, tenascin-C and plasminogen activator inhibitor-1. Down-regulated transcripts included matrix metalloproteinase-1 and thrombomodulin. Considering the pre-existing knowledge of these gene products, the authors went on to discuss the potential relevance to vascular disease of this, rather restricted, alteration in gene expression.

2.2. Genetic polymorphisms

Novel drug targets can also be identified through the analysis of genetic polymorphisms. In the following section, we discuss how the association of a particular genotype with a particular phenotype could be used to define risk or maximise drug

benefit. It should also be recognised that polymorphisms can also provide clues as to which genes or gene products would best be targeted. However, determining whether individual allelic variants are of therapeutic relevance is likely to require considerable further work [13].

2.3. Orphan receptors and reverse pharmacology

The identification of novel sequences of putative receptors can be achieved using computational methods, such as searching EST databases, but the ability to similarly identify the natural ligands to these receptors is not possible by these methods. Putative receptors identified by gene cloning that exhibit homology to known receptors in the existing superfamilies, but for which no known ligands have been identified, are referred to as orphan receptors [14].

Ligands for orphan receptors can be identified using 'reverse' molecular pharmacology, whereby receptors are recombinantly expressed in functional assays to search for the associated ligand. Screening may begin with compounds that are already characterised as ligands of receptors of that particular family. Subsequently, novel activating ligands can be screened for, using compound libraries, tissues, biological fluids and cell supernatants. Thus, selective agonists and antagonists can be defined, practically disregarding their physiological or pharmacological role, which are then tested for potential beneficial effects.

With more than 1000 receptors identified to date (http://www.gpcr.org/7tm/), the superfamily of the G protein-coupled receptors (GPCRs) is one of the largest known gene families. GPCRs are found in a wide variety of cells and respond to a vast range of agents [15]. They are of particular interest because they have an excellent history as being drug targets; examples of GPCRs already targeted include the α- and β-adrenoceptors, and the angiotensin II and dopamine receptors. Ligands for many of these integral plasma membrane receptors remain to be identified [16].

After identifying a human gene encoding a novel GPCR with a 75% sequence similarity to the rat GPR14 orphan receptor, Ames et al. [17] used cells expressing the human GPR14 to screen 700 potential agonists for the activating ligand. Urotensin II (UII) was identified as a potent stimulator of Ca^{2+} responses in these cells. They also showed that UII caused marked increases in peripheral resistance and severely depressed myocardial activity in monkeys. Although recently identified in humans [18], UII was isolated from fish more than 20 years ago. The recent identification of UII as the ligand of the human GPR14 receptor by reverse pharmacology will finally accelerate research efforts into this peptide, which, because of its vasoactive actions, might play a role in hypertension or heart failure.

2.4. Forward genetics

A further method of identifying gene products which have potential as drug targets is to randomly mutate the genome, select mutants with interesting defects and then define the gene involved [19]. So far, this method of 'forward' genetics has been mainly used to investigate signalling pathways, such as those involving the interferons.

3. Genetic polymorphism

Elucidating the influence of DNA sequence variability within genes holds tremendous potential both for understanding the genetic basis for complex disorders and improving the ways in which we treat them. The most abundant type of DNA sequence variation in the human genome are single nucleotide polymorphisms (SNPs) [20]. Between two individuals, there may be several million single base pair differences, accounting for about 100,000 amino acid differences between their total complement of proteins. If SNPs are located within coding regions, they may or may not change the amino acid sequence; if they do, this might, or might not, alter protein function. However, most SNPs are located outside protein-coding regions. These are also important because they may be used to map gene variants associated with particular phenotypes, or may be crucial for mRNA stability or rate of transcription. There are a number of projects creating libraries of SNPs, many of which are available publicly and free of charge [21]. The SNP Consortium, a nonprofit grouping of drug companies, information-processing companies, academic institutions and the Wellcome Trust, is currently creating a high-density SNP map with 200,000–300,000 SNPs (http://snp.cshl.org/).

Unravelling the variation in the coding sequence of genes that contribute to complex multifactorial polygenic conditions, such as hypertension, will enable these conditions to be redefined in molecular terms. Thus, no longer will 95% of those patients with hypertension be labelled as primary hypertensives. Instead, they will be subgrouped according to their DNA profile. This is likely to influence subsequent mortality or morbidity, as well as the most appropriate treatment for a given individual, and how aggressive that treatment should be. Such information will not only be valuable for the treatment of established disease but will also allow for population screening for diseases in the pre-symptomatic stage. Thus, we might be able to predict who will be most likely to develop hypertension. Subsequent action, be it, for example, more frequent blood pressure measurements, lifestyle advice or pharmacological intervention, might itself depend on the genetic profile. Despite the potential medical benefits of using genomic data to evaluate risk, there are significant ethical issues concerning the use of this information. Patients might be uneasy that such detailed genetic information about them exists, or indeed, might not want to know how and when they are likely to fall ill. Whether this information should be available to insurance companies or government bodies is a further major issue that will have to be resolved.

3.1. The genetic basis of hypertension

The relationship between genetic variation and hypertension has been the focus of much interest [22,23]. Despite knowledge of numerous proteins involved in blood pressure homeostasis, to date, no genotype has been proven to be causally related to primary hypertension [24]. Although some investigators find an association between certain polymorphisms and the presence of hypertension, these have not always been corroborated in other studies. A number of factors may be important in understanding inconsistencies in the relationships between polymorphisms and disease. For example, different populations, such as those with different ethnic backgrounds, might have

different patterns of polymorphisms which are relevant to a given condition. Thus, the population being studied and therefore, the population to which the results will be applicable, should be characterised as precisely as possible. Care should be taken to ensure that, when investigating the effect of polymorphisms on a given phenotype, controls are selected from the same population as the patients. Furthermore, such studies may be subject to publication bias. For example, the angiotensin converting enzyme (ACE) gene is characterised by a polymorphism based on the presence (insertion, I) or absence (deletion, D) of a 287-base pair *alu* repeat sequence. A meta-analysis investigating the association of this I/D polymorphism with myocardial infarction found that smaller studies were more likely to be published if they showed a significant effect [25] (see Fig. 1).

Attempts are often made to find associations between individual polymorphisms and a given phenotype. Recently however, limitations of this strategy have been emphasised. For example, the ACE gene D/D genotype is clearly linked with increased circulating levels of the enzyme, but the association between genotype and cardiovascular disease is inconsistent [26–32]. Rieder et al. [33] examined the entire ACE gene from 11 individuals. In all, 78 varying sites were identified, which resolved into 13 distinct haplotypes. A number of the variant sites were found to be in absolute linkage disequilibrium with the I/D polymorphism, so that, with reference to this, haplotypes could be sorted into two distinct groups. In addition, a major subdivision of the D group was identified which should allow more informative analysis of the cardiovascular traits associated with this variation. The consideration of non-coding regions in this study was also shown to be of importance as the majority of site variants in absolute disequilibrium with the I/D polymorphism were found in these regions and would have been missed if only coding regions had been examined.

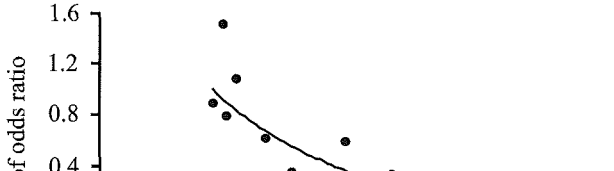

Fig. 1. Funnel plot to investigate publication bias in studies examining the link between the ACE I/D genotype and the risk of myocardial infarction. Estimate of the odds ratio for DD vs. ID/II genotypes against trial size (log scale) for each study is shown. The horizontal dashed line shows the pooled estimate of the log odds ratio across all studies. In the absence of publication bias, studies of all sizes would be expected to be scattered equally above and below the pooled estimate, with smaller studies having larger standard errors, creating a horizontal funnel effect. However, the smaller studies are almost all above the pooled estimate line. This is consistent with publication bias, where smaller studies are only published if they show significant effects. Adapted from Ref. [25] with the kind permission of Dr. O'Toole.

A lack of appreciation of the effects of gene–gene interactions may also help to explain the inconsistent results regarding the genetic analysis of hypertension. Williams et al. [34] recently addressed this by examining the effects of interactions of alleles at four candidate loci, three of which were in the renin–angiotensin system (angiotensinogen, ACE and angiotensin II type 1 receptor). A total of seven polymorphic sites were examined in hypertensives and age-matched normotensive controls. There were no significant effects of allele or genotype frequencies at any one locus. Thus, no one site alone showed any significant association with hypertension. However, in order to examine the effects of the interactions between multiple loci, 120 multilocus comparisons were made for each group. In hypertensives, 16 of these comparisons were found to be in significant linkage disequilibrium, whereas none were found in normotensive subjects. This result indicated that genetic interactions between multiple loci, rather than variants of a single gene, were important to the genetic basis of hypertension.

3.2. The risk of hypertensive complications

The association of disease with genetic polymorphism can be taken a step further by linking genetic variation with the consequences or complications of a given condition. Thus, in the case of hypertension, we might be able to define genetic patterns that determine blood pressure but might also be able to elucidate polymorphisms that influence the subsequent development of left ventricular hypertrophy (LVH), heart failure, myocardial infarction, or stroke. Such information would enable better stratification of the risks of morbidity and mortality in different patients and aid the targeting of therapy to higher risk groups. A recent meta-analysis has been published [35], investigating the effect of the I/D polymorphism of the ACE gene on the development of LVH in hypertensives, and showing that in untreated hypertensives, the D allele behaved as a marker for LVH. Indeed, those untreated hypertensives with the DD genotype had around twice the risk of developing LVH compared to those with the II genotype.

3.3. Individualised drug therapy

Genetic heterogeneity accounts for significant variation between individuals in their responses to drugs. Complex genetic factors influence beneficial and adverse reactions, as well as drug metabolism and disposition [13,36]. Thus, some drugs may be either more efficacious or more toxic in certain groups of patients than in others. At the molecular level, genetic heterogeneity may be responsible for subtle conformational changes at the drug's binding site, for differences in the cellular uptake or removal of a drug, the precise physiological function of the drug target, or even for interaction of the drug with molecules other than the primary target.

Currently, physicians have to make empirical decisions when choosing a drug and at what dosage to give it, using information that has been gathered on the basis of population averages, rather than individual profiles [37]. The division of patient populations into smaller subgroups according to their genetic profiles should, ultimately, allow for a more informed and rational prescribing strategy [38]. It should be possible to predict, not only the most appropriate drug, but also the dose required. This will represent a highly

significant advance, especially in diseases in which it can take months or years of treatment to observe whether a positive response has occurred. It should also help to address the problem of demand outstripping limited healthcare resources by limiting the use of unnecessary drugs, ineffective drugs and of drug-related side effects [39]. It should also be noted that prescribing decisions could possibly be based simply on the association of drug response with a given haplotype, and that knowledge of gene function should not be required.

These principles apply not only to currently prescribed and future drugs, but may also be applied to previously failed drugs. Thus, an alternative indication for a drug or a responsive subgroup in an otherwise unresponsive population might be identified. The potential to 'rescue' drugs may be particularly relevant to those in which it has been difficult to strike a balance between efficacy and toxicity in the general population. Currently, new indications for a marketed drug are often discovered by physicians by trial-and-error; genetic data could provide a more informed way of rapidly identifying several of those indications.

There are concerns regarding the development of individualised drug therapy. When a drug has been shown to be of particular benefit to a well-defined population, the pressure to prescribe it in these circumstances will be significant. Indeed, it might be considered negligent if it were not so prescribed. Should the patient be inadequately or inaccurately genotyped, they might not receive the most appropriate medication. Indeed, they may receive a drug that is harmful to them. In clinical practice, the treatment of a condition such as hypertension is relatively simple for most patients. This might no longer be the case if every patient has to undergo genotyping, with subsequent prescribing decisions based on the results. There will be implications for the continued education of doctors, the cost of introducing new diagnostics, as well as for consultation time with patients, who will require counselling both before and after genotype testing. There is also a concern that large pharmaceutical companies might view the development of individualised therapy negatively, given that it has the potential to exclude patients and shrink markets. Whereas we have eluded to the potential problem for doctors concerning the pressure to prescribe a drug shown to be of benefit in a well-defined group of patients, for pharmaceutical companies, this would improve market share for a given product. Indeed, a better drug at the outset would be more resistant to later competing products.

3.4. Hypertension and individualised prescribing

It is well-established that there is considerable variability in the response of individuals to different anti-hypertensive drugs [40]. This appears to reflect different mechanisms of blood pressure elevation. Provided that associations can be made between specific genetic patterns and the therapeutic response to given drugs, more efficient use of antihypertensive agents should be possible, based on an individual's genetic profile. The principle of targeting drug therapy to subgroups of patients with hypertension can be illustrated by the treatment of two single gene disorders, Liddle's syndrome and glucocorticoid-remedial aldosteronism.

Liddle's syndrome is an autosomal dominant condition characterised by hypertension, hypokalaemia and suppressed renin and aldosterone levels due to volume expansion.

Blood pressure is very sensitive to both a low sodium diet, as well as triamterene and amiloride treatment. A number of different mutations of either the β-subunit or the γ-subunit of the epithelial sodium channel (ENaC) have been characterised that result in the Liddle's syndrome phenotype [41–46]. ENaC mediates the electrogenic entry of sodium ions from the lumen into the cell; in the collecting duct of the kidney, it is the last site for sodium reabsorption and one of the most important [47]. The ENaC mutations result in a reduction in intracellular turnover of the channel and an increase in the number of active channels exposed at the cell surface. This leads to increased ENaC activity in the distal renal tubule with excess sodium reabsorption. ENaC is selectively blocked by amiloride and triamterene, explaining the high degree of efficacy of these drugs in treating the syndrome.

Glucocorticoid-remedial aldosteronism (GRA) is another autosomal dominant single gene disorder, characterised by hypertension with variable hyperaldosteronism and high levels of other abnormal mineralocorticoids. The genetic basis for this condition is a novel gene on chromosome 8, which represents duplication arising from unequal crossover, fusing the regulatory region of 11β-hydroxylase with the coding region of aldosterone synthase. Adrenocorticotrophic hormone (ACTH), rather than angiotensin II, therefore controls the expression of, and leads to excessive production of, aldosterone. GRA can be treated with glucocorticoid, to suppress ACTH secretion, and thus, the expression of the abnormal gene.

Of course, blood pressure in the majority of primary hypertensives is likely to be determined not only by multiple genes, but also by environmental factors. However, despite the conflicting data concerning polymorphisms and the risk of developing hypertension, there are reports indicating differing responses to antihypertensive medications. Having found an association of the A^{1166C} polymorphism of the angiotensin II type 1 receptor (AT_1-R) with arterial stiffness, measured by carotid-femoral pulse wave velocity (PWV), in hypertensive individuals, Benetos et al. [48] went on to examine its influence on aortic stiffness after treatment with the ACE inhibitor, perindopril, and the calcium channel blocker, nitrendipine. With perindopril, there was a significantly greater reduction in the PWV in carriers of the C allele than in those without this allele. With nitrendipine, the observed effect was reversed. Thus, there was a greater reduction in PWV in those without the C allele than those with it. The authors concluded that, because of previous evidence linking higher PWV with increased cardiovascular risk, it might be useful to consider the AT_1-R genotype when prescribing an ACE inhibitor or calcium channel blocker for a hypertensive individual. Interestingly, an earlier study had linked a polymorphism in the angiotensinogen gene to differences in the blood pressure response with ACE inhibitor therapy in previously untreated primary hypertensives [49].

The β-adrenoceptor (βAR)-G_s protein is essential to the activation of adenyl cyclase in cardiac and vascular smooth muscle cells and has been targeted for investigation of hypertension. Indeed, a common polymorphism in the sequence encoding the α-subunit was found to be associated with blood pressure in hypertensive individuals [50]. In the same study, the blood pressure response to β-blockade was significantly associated with the presence or absence of this polymorphism. A recent study by Cockcroft et al. [51] showed that two common polymorphisms of the β_2-adrenergic receptor influence both

arterial and venous vascular reactivity to the β-agonist, isoproterenol, in male normotensives. It can be speculated that these polymorphisms might influence both the genesis of hypertension and the response to therapy. A recent study looking at the effect of SNPs of the β_2-adrenergic receptor on the response to β-agonists in asthmatics illustrated the importance of investigating the interactions of multiple polymorphisms in evaluating population variation in drug response [52]. Thus, whereas individual SNPs were poorly predictive of bronchodilator response, haplotype pairs were significantly predictive. Furthermore, when two of the haplotypes were expressed in cultured cells, there was an approximately 50% greater receptor mRNA expression and receptor density in those cells transfected with the haplotype associated with a greater physiologic response compared to those transfected with the lower response haplotype.

3.5. Clinical trials

Genotype data will also be of benefit to drug development in clinical trials. Thus, in phase I studies, genotype could be correlated with both efficacy and adverse events. This information could be refined in phase II trials, so that phase III trials could be performed on an identified subgroup of patients who are more likely to respond well to the drug and to exhibit fewer side effects. Thus, not only could clinical trials be smaller, faster and more efficient, but the resultant drugs would be expected to have improved efficacy and a better safety profile. It may be that a drug would prove to be suitable only for a rather small number of patients, such that it would be uneconomical for a drug company to pursue. In this case, the risks of embarking on expensive larger trials could be minimised, with obvious overall cost benefits. However, whether sufficient genomic information could be gathered in early clinical studies to help determine the most appropriate subjects to recruit to phase III trials or, indeed, to determine whether such studies should be performed at all, remains to be seen. This information might be more useful in larger post-marketing studies to further characterise the features of the population to which the drug is most suited. However, at this stage, revealing the market for a particular drug to be significantly smaller than expected could be financially disastrous. Despite this, it should also be possible to retrospectively genotype stored blood samples in an attempt to explain equivocal results or 'outliers' in phase III trials [39].

4. Bioinformatics

There has been an explosion of data arising as a result of the genomic revolution. In order to fully exploit this information, so that valid drug targets can be efficiently defined, it is critical that the data from all sources is integrated intelligently. Bioinformatics involves the application of information technology to the management and analysis of biological data. It is a cross-disciplinary activity, incorporating aspects of computer science, software engineering, mathematics and molecular biology [53]. Although necessary, the development of effective bioinformatic systems represents a significant challenge.

5. Conclusion

The understanding and redefinition of disease in molecular terms will fundamentally change the way in which drugs are discovered, developed and used in the clinic. The ability to more accurately quantify risk of disease, along with the potential for new drugs, and more appropriate use of drugs in individuals will allow for greater confidence among clinicians that they are doing the best for their patients. However, the implications of these developments might not all be positive. For example, the overall implication for healthcare costs is not known and there are a number of ethical issues which must be resolved.

Appendix A. Discussion

M. Reidenberg: I have to respond to your statement that negative studies can't get published. The studies of this have shown that in general, "negative studies" don't get published because the scientists don't bother writing them up and submitting them. The scientists are not prepared to put additional time into what they perceive as a negative study. Speaking personally for Clinical Pharmacology and Therapeutics, if the hypothesis was reasonable and the negative study is reasonably definitively negative, then we'll always accept a "negative study".

P. Joubert: With our current knowledge, the notion of smaller, faster, more efficient trials remains naive. Drug development to me is a risk–benefit analysis, and you can, I guess, assess a rapid onset benefit in a very short, fast trial, if you have a very highly responsive group. If it is long-term therapy however, you need a reasonable number of patients exposed to get a handle on the safety. The worst scenario can be to come up with a therapy in a small very responsive group to find out that there is a one-in-a-thousand very severe, or long-term adverse event you cannot deal with. From that point of view, I still have some reservations, maybe our position will change with time. The other problem that I see is that even the good cases that I quoted—one that is often shown is the association between the APO E gene expression and Alzheimer's disease. If you look at the responders versus the non-responders, the non-responders still do better on drug than on placebo. So there is still a percentage of patients responding. The genotype, at present, just gives you a likelihood within a population but it does not give you the exact chance of the individual. The group with a low chance of responding still includes responders. So, I think that it remains very tough to sort out. It is perhaps just our lack of understanding of the polygenic nature of some of the things we think are monogenic.

D. Webb: I agree with all you said, and, of course, it is possible to lower blood pressure in healthy people with almost any vasodilator drug, and we know that lowering blood pressure will reduce stroke risk at all levels of pressure. So there are benefits to be gained in all subjects. I suppose, for the moment, I'm expecting these sorts of data to come from late phase IV studies, such as, say the big trials, like Survival and Ventricular Enlargement (SAVE) and Vasodilator-Heart Failure Trial (V-HeFT) studies. Here, genotyping has already been done, and one can trawl through and generate new hypotheses that can be tested independently in other large studies. This probably has little to do with the drug industry, but may ultimately rationalise use of drug treatment by the clinician. In terms of

small, fast, and efficient studies, I'm still waiting for someone to show me exactly how that's going to work. That may happen in a small number of specific diseases, but probably not in the major complex cardiovascular disorders.

G. Levy: I can see where genotyping can help in selecting the best drug for the particular subtype of a disease. My question is, can genotyping help in selecting the best dose of a drug for therapy? I was thinking from a pharmacodynamic point of view, in other words, predicting the appropriate concentration of the drug for an individual patient, and then, of course, we have the drug metabolism part of it, but I'm now focussing on the dynamic.

D. Webb: Obviously, if there is a well-recognised adverse effect that is dynamically related, then one might use lower doses in subjects whom one would predict genetically to be at risk. There's the well-known J-shaped curve for blood pressure lowering; one perhaps does not want to lower blood pressure more than a certain degree, and it might be possible to avoid a particular genotype to prevent excessive blood pressure lowering in some cases. I think genotyping is much more likely to be useful to predict the best drug, as opposed to the dose at which it might be used.

A. Breckenridge: You make less than a compelling case for following a molecular biology approach to the discovery of new anti-hypertensives compared to, let's say for example, the wealth of information that's coming out of the Heart Outcomes Prevention Evaluation (HOPE) study and clinical observation; would you agree with that?

D. Webb: Yes, in part. I didn't cite any of his studies, but Morris Brown is taking a number of interesting approaches to this problem and one is a rotating treatment study for individual untreated hypertensives. He takes them through a range of the four main classes of drugs used in the UK (diuretic, beta-blocker, calcium-antagonist, ACE inhibitor) and he finds individual differences in responsiveness. His study is still too small at present to pull out key genes, but that approach, particularly if other groups took it on with him, would be a way to understand better the drugs we are currently using. I think there is a lot of utility in improving our use of the currently existing drugs, at least from the point of view of the patient. That approach is worthwhile and we need to draw on different phenotypes; I'm very interested in the role of sodium loading and exercise, and their effects on blood pressure, and whether critical phenotypes that link to genotype might emerge from those sorts of approaches. Of course, those who are interested in cardiovascular physiology will know that the peripheral blood pressure at the brachial artery is not what the heart sees, and central blood pressure can be very different between individuals with the same brachial pressure. So, we already know the most common phenotype is probably wrong.

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