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How the practice of allergy shows the promise and challenge of personalized medicine

Sean A. McGhee*

Department of Pediatrics, Immunology and Allergy, 269 Campus Drive, CCSR Bldg Rm 2115, Stanford, CA 94305, USA

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ABSTRACT

Personalized medicine seeks to stratify therapies according to individual characteristics, and by so doing improve effectiveness and reduce complications. However, there are not many models of care that is highly stratified within a single diagnosis in this manner. One potential model is the practice of allergy, in which care is tailored to specific allergens for individual patients within the broader context of care for rhinitis or asthma. Allergists have already confronted many of the same regulatory issues anticipated for personalized medicine. The history of allergy practice also anticipates some of the patient safety concerns that may arise from tracking and using highly personalized medical information. Finally, the therapy of allergy and asthma has been at the forefront of attempts to incorporate pharmacogenomics information into patient care. Individualized therapy has always been central to the practice of allergy, and so provides a useful proving ground for personalized medicine as a concept of care.

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In some sense, medicine has always sought to provide care in the model of personalized medicine. We identify the features in each individual that suggest one diagnosis and not the other, and, in so doing, avoid treating the type I diabetic as a type II diabetic, despite the surface similarities of the disorders. Personalized medicine simply takes this to a different extreme, providing stratification of therapies to patients with the same diagnosis, recognizing that a disease with similar pathophysiology may not manifest identically in different individuals.

The human genome project provides a technical framework, that of genetic and heritable variability, on which to base at least some of this stratification. For example, algorithmic dosing based on single nucleotide polymorphisms (SNPs) and other personalized factors provides rapid and accurate dosing of warfarin [1]. Targeted therapy of oncologic disease, such as with Herceptin and Gleevec improves outcomes and reduces side effects of therapy [2–4]. There have been concerns about the challenges and cost of implementing personalized therapies though [5]. Models of how such care has been implemented in the past would be helpful.

Since at least 1911, allergists have been providing exactly this kind of stratified care, and so have already confronted and successfully managed many of the logistical, regulatory and technical barriers that have limited the introduction of stratified care in other fields [6,7]. This has happened because the practice of allergy identifies a unifying diagnosis for the patient's condition, such as allergic rhinitis or

asthma, but tailors therapy to the specific allergens that are triggering episodes of the disease. Each patient presenting for care of atopic disease is systematically assessed, by history and by skin testing or specific IgE testing, to determine the specific profile of antigens that trigger disease. In many cases, a specific therapeutic intervention, immunotherapy or desensitization, that complements more generic medical care for the asthma or rhinitis will be performed based on this testing. This therapy is almost always designed specifically for the individual patient and is custom-prepared and delivered on an individualized schedule [8]. While other fields, especially oncology, have made enormous strides in targeting therapy, few other than allergy have been performing such targeting since 1911.

There are several challenges that arise from providing such stratified care within the current medical system. These include problems in demonstrating the efficacy of therapy when the same therapy differs between individual patients, and regulatory challenges in managing the expectations of having single, unique indications for tests and therapies. There are also significant patient safety concerns related to managing the large information load necessary to ensure that each patient receives accurate therapy. Finally, there are ongoing questions and challenges in defining the correct phenotypes within which to stratify care.

First, personalized medicine may confront problems with demonstrating the efficacy of interventions that are highly tailored. Regulatory approval of therapies requires that the efficacy of a therapy be demonstrated, usually in large, randomized, placebo-controlled trials. However, immunotherapy and other highly stratified therapies may be better thought of as a concept of care, a basic principle which can be implemented in a number of different ways.

* Fax: +1 650 498 6077.

E-mail address: smcghee@stanford.edu.

For immunotherapy, a set of several different antigens is created based on an individual profile of sensitivities and this unique mixture is provided to induce tolerance to these antigens in the patient. Regulatory agencies might be reluctant to accept proof of efficacy for this general concept of care, preferring to certify a specific drug for a specific indication. This is reasonable because it avoids the problem of having nebulous boundaries surrounding what is or is not an approved therapy. However, this same desirable goal inhibits the growth of highly tailored therapies by requiring each to be individually approved, dramatically increasing the cost of regulatory approval.

Currently, in the US, production of each antigen extract is approved by the Center for Biologics Evaluation and Research (CBER) at the Food and Drug Administration (FDA) [9]. However, the mixing and administration of the antigens is performed by the individual allergist. This means that although there is substantial regulatory authority over the extract's production, its use and implementation is less centrally regulated, primarily through physician licensure, hospital privileges and board certification. Maintenance of Certification procedures for allergists have been the primary mode by which ongoing quality assurance has been implemented, and these programs are young and their effectiveness as of yet is uncertain [10–12].

Similar issues will arise for other tailored and stratified therapies within personalized medicine. The most appropriate venue for regulatory oversight of personalized therapies should be determined carefully and an active decision made about the best level for regulated personalized medicine practices. For example, the FDA changed the labeling of warfarin to include the general concept that genetic information should be considered when planning dosing. Many felt this was premature as such recommendations had not been incorporated in professional guidelines at the time, a clear example of conflict over the appropriate level of regulation [6]. It will be important to continue to assess who should maintain supervision and oversight of the application of personalized medicine.

The second consideration, patient safety, has proven to be a significant concern in the practice of allergy, particularly with immunotherapy. Patient safety in personalized medicine has typically been thought of in positive terms, because of its potential for predicting adverse drug reactions and other poor outcomes. But there is another side to personalization of therapy. Allergy immunotherapy has the potential for fatal side effects from anaphylaxis, and it has been demonstrated that one of the most common scenarios (approximately 25%) in which anaphylaxis results is a wrong drug, wrong dose, or wrong patient error [13]. A busy allergy practice may have hundreds of patients, each on their own specific immunotherapy, receiving injections on a weekly basis, and with dosing which changes with each visit. Accurately managing this information load is challenging, and is the focus of high profile patient safety efforts from the major allergy societies. Many interventions that are now routine in allergy practices, such as systematic identification of high-risk patients, multiple level checks of dosing and patient identification, use of photographs in medical records, and expansion of electronic medical records, may need to be considered carefully as more highly stratified therapies become a reality in other fields.

The information problem will be compounded as patients with multiple medical problems receive multiple highly stratified therapies. Already there is difficulty in managing drug interactions and polypharmacy in the elderly and patients with multiple medical problems [14]. These problems become significantly more intense when therapies are highly tailored. It may be difficult to predict specific drug interactions outside of those that may be expected based on drug class. For example, there are known interactions between immunotherapy and beta blockers or ACE inhibitors, since these therapies inhibit the response to anaphylactic events and promote the likelihood of anaphylaxis resistant to therapy [15].

However, there is almost nothing known about interactions between specific antigens and other pharmaceuticals, or even whether such interactions exist. Highly tailored therapies are also susceptible to intrinsic drug interactions as well. The mixing of antigens is complex and many antigens will change the potency of others when mixed [8]. Hence an additional set of potential errors outside of wrong drug errors can occur when a therapy must be individually tailored. Instead of just a dosing error, there exists the possibility of an error in compounding the therapy itself, with a much greater likelihood than occurs in standard, single drug therapy.

There are no simple solutions to these issues. As therapies become more stratified, the base of patients on which to conduct trials becomes smaller, while the potential for interactions expands exponentially. There is no conceivable hope of conducting blinded randomized trials of all possible combinations. There will have to be a much greater reliance on electronic records, data mining, and more advanced statistical methodology to adequately monitor for such effects, followed by more rigorous trials as necessary.

A further challenge in providing highly stratified therapy is in defining the correct phenotypes in which to stratify. The broadest category in allergy is the notion of atopy, the general predisposition to develop a T_H2 -mediated, allergic response to specific, generally insignificant antigens that do not represent a threat to self (pollens, insects, animal dander). However, many phenotypes have at least two sub-phenotypes, one due to atopy and one that is due to other causes. Both rhinitis and asthma, for example, may be due to atopic or non-atopic causes. If not correctly assigned to an atopic cause, providing highly stratified therapy against specific allergic sensitization does nothing to improve the patient's condition, and places them at risk for anaphylaxis. Currently, there is an assumption made in most cases that if a patient has rhinitis or asthma and is atopic, that the atopy must be contributing at least in part. However, this is difficult to prove, and a lack of response to allergen immunotherapy alone might be the only way to demonstrate the profound role that non-atopic causes might be playing. It would spare much cost, time and effort if that could be adequately known ahead of time, but usually it is not. Making a correct diagnosis remains the most critical part of a patient's therapy, and in some cases our ability to provide highly stratified therapies may outstrip our ability to correctly diagnose patients.

The response to this issue is clear. While personalized medicine provides exciting possibilities and could significantly improve the care of patients, it is critical to recognize that the job in "non-personalized medicine" is not yet done. There remains a need for epidemiologically based studies defining phenotypes outside of highly stratified care, since stratified therapies are based on arriving at correct phenotypes in the first place.

While allergen immunotherapy has been around for a century, the field of allergy has also moved into areas of stratification more typically associated with personalized medicine. Oncology and warfarin dosing have led the way in incorporating pharmacogenetics into medical practice, but there has also been considerable work done in asthma therapy [16]. For many years, there has been debate about the most appropriate step-up therapy to use for asthma. While many people respond to low doses of inhaled corticosteroids, many do not, and these people require some type of step-up therapy. There is an abundance of options for step-up, including a leukotriene inhibitor, doubling the dose of corticosteroid, adding a long-acting beta agonist, or using a long-acting anticholinergic agent. However, despite several well-designed and appropriately powered clinical trials, there is no consistent answer as to which is the best strategy [17–19]. It appears that different people respond differently, and that current understanding of asthma does not permit a prediction about who will respond to which therapy.

The best understood pharmacogenetic stratification has been the use of beta-adrenergic receptor polymorphisms [16]. The beta-adrenergic receptor is produced from a single exon gene with at

least 49 known polymorphisms [20,21]. Three of these have been well studied, the Arg16Gly (ADRB2+46G>A), Gln27Glu (ADRB2+79C>G) and Thr164Ile (ADRB2+491C>T). These were shown to vary with ethnicity (as asthma disease burden does), and do have functional consequences for the beta-adrenergic receptor [22–24]. Beta agonists play a vital role in rescuing asthmatics with severe exacerbations, but overuse in some individuals is associated with poor outcomes and fatalities. Since overuse of beta agonists in asthma therapy is associated with poor outcomes, there was much hope that such polymorphisms could predict those who would have poor response to beta agonist therapy [25–27]. Considerable effort has been placed into the search for polymorphisms in this gene that would predict response to therapy, with complex and confusing results [28,29]. It remains unclear whether ADRB2 polymorphisms are useful in practice.

However, the beta-adrenergic receptor is not the only attempt to stratify asthma care and provide targeted therapies. A somewhat more successful stratification has been due to the realization that aspirin-sensitive patients with asthma (aspirin exacerbated respiratory disease, or AERD) have a unique disorder with a unique response to targeted therapy [30–32]. Aspirin-sensitive patients appear to be particularly sensitive to leukotrienes, and highly effective blockade of the leukotriene pathway, such as with zileuton, a leukotriene synthesis blocker, improves their care [31,33]. Furthermore, these patients are responsive to aspirin desensitization, with marked improvement in asthma and sinus disease when appropriately desensitized [34–37]. For these patients, these targeted therapies are truly life-changing.

These two situations demonstrate both the promise and challenge of personalizing medicine. Such stratification promises to revolutionize asthma care, which is likely a final common pathway for multiple etiologies. Personalized medicine will play a vital role in effectively treating asthmatics in the future. But it is not enough to know the sequence of the genome and the polymorphisms therein to be able to apply predictive and personalized therapy. We must be able to identify which polymorphisms are predictive out of the many that are irrelevant, and the underlying mechanism of their action must be simple enough that they can be incorporated into clinical care algorithms without undue complexity. We must also be able to handle the increased information load from tracking and managing patient descriptors that allow us to make these predictions. And finally, the cost of implementing this stratification must be worth the benefit derived from it.

That SNP or other polymorphisms will predict response to therapy in a simple or easily understood fashion is not supported by our current experience. Most common attributes are polygenic, currently unable to be adequately modeled even in a research setting, let alone a clinic, and there remain large amounts of unexplained heritable variability in traits, such as variations in transcription due to RNA processing [38]. It is reasonably safe to assume that when finally adequately explained, the predictive models will not be simple. This will then entail a reliance on computer-assisted diagnosis, tracking and management that is currently unprecedented. Ultimately, this may be a good thing for medicine, reducing error in the same way that automating pilot decisions in aircraft improves safety and reliability, despite pilots' initial reluctance to let go of the controls. However, electronic medical records are a long way from approaching the safety and reliability of even the simplest of autopilots, and the mechanisms of living systems easily outstrip the complexity of our most advanced aircraft.

Finally, the incremental costs of testing, tracking and managing stratifiers must be worth the incremental gains in healthy life that result. Comparative effectiveness research has clearly demonstrated that interventions that improve health, like all human activities and products, come at widely varying costs, from interventions that are cost-saving to those that cost well over \$1 million for every year of

healthy life saved per person [39]. Stratified therapies will also likely run this entire gamut, with some being highly effective and cost-saving, while others may be expensive and barely effective. Assessment of the comparative effectiveness of stratified therapies is reasonable and an important investment [40]. As long as attention is paid to this question, there is no intrinsic reason why personalized medicine should not be as equally or more cost-effective than current therapeutic strategies.

For almost exactly 100 years, allergists have been implementing personalized and highly stratified therapies for patients. They have been at the forefront of the scientific effort to incorporate data from the human genome project into clinical care. They have confronted many of the regulatory and patient safety challenges that arise out of delivering highly personalized therapy. But even after 100 years, we are still learning how to do this effectively and safely. Personalized medicine will not arrive one day, but will be won by inches, through consistent and careful application of science.

References

- [1] U.I. Schwarz, M.D. Ritchie, Y. Bradford, C. Li, S.M. Dudek, A. Frye-Anderson, et al., Genetic determinants of response to warfarin during initial anticoagulation, *N. Engl. J. Med.* 358 (10) (Mar 6 2008) 999–1008 [Research Support, N.I.H., Extramural].
- [2] L. Yan, R. Beckman, Pharmacogenetics and pharmacogenomics in oncology therapeutic antibody development, *Biotechniques* 39 (Suppl. 10) (2005 Oct) S565–S568 Review.
- [3] M.J. Duffy, J. Crown, A personalized approach to cancer treatment: how biomarkers can help, *Clin. Chem.* 54 (11) (Nov 2008) 1770–1779 [Research Support, Non-U.S. Gov't Review].
- [4] I. Collins, P. Workman, New approaches to molecular cancer therapeutics, *Nat. Chem. Biol.* 2 (12) (2006 Dec) 689–700 [Research Support, Non-U.S. Gov't Review].
- [5] E.B. Elkin, D.A. Marshall, N.A. Kulin, I.L. Ferrusi, M.J. Hassett, U. Ladabaum, et al., Economic evaluation of targeted cancer interventions: critical review and recommendations, *Genet. Med.* (Jun 1 2011). [Epub ahead of print].
- [6] R. Conti, D.L. Veenstra, K. Armstrong, L.J. Lesko, S.D. Grosse, Personalized medicine and genomics: challenges and opportunities in assessing effectiveness, cost-effectiveness, and future research priorities, *Med. Decis. Making.* 30 (3) (May-Jun 2010) 328–340.
- [7] L. Noon, Prophylactic inoculation against hay fever, *Lancet* 1 (1911) 1572–1573.
- [8] L. Cox, H. Nelson, R. Lockey, C. Calabria, T. Chacko, I. Finegold, et al., Allergen immunotherapy: a practice parameter third update, *J. Allergy Clin. Immunol.* 127 (Suppl. 1) (Jan 2011) S1–S55 [Practice Guideline].
- [9] Allergenic. U.S. Food and Drug Administration, 2011 [cited 2011 June 16, 2011]; Available from: <http://www.fda.gov/BiologicsBloodVaccines/Allergenic/default.htm>.
- [10] J.M. James, M. Corbett, The American Board of Allergy and Immunology maintenance of certification program, *Ann. Allergy. Asthma Immunol.* 106 (6) (Jun 2011) 549 [Letter].
- [11] J.M. James, M. Corbett, The American Board of Allergy and Immunology maintenance of certification program: “to do or not to do? That is the question.”, *Ann. Allergy Asthma Immunol.* 105 (6) (Dec 2010) 485–488.
- [12] M.S. Dubravec, The burdensome maintenance of certification process, *Ann. Allergy Asthma Immunol.* 106 (4) (Apr 2011) 351 [Letter].
- [13] D.I. Bernstein, T. Epstein, Systemic reactions to subcutaneous allergen immunotherapy, *Immunol. Allergy Clin. North Am.* 31 (2) (May 2011) 241–249 [viii–ix].
- [14] J.J. Lopez-Picazo, J.C. Ruiz, J.F. Sanchez, A. Ariza, B. Aguilera, D. Lazaro, et al., Prevalence and typology of potential drug interactions occurring in primary care patients, *Eur. J. Gen. Pract.* 16 (2) (2010 Jun) 92–99.
- [15] M.J. Hepner, D.R. Ownby, J.A. Anderson, M.S. Rowe, D. Sears-Ewald, E.B. Brown, Risk of systemic reactions in patients taking beta-blocker drugs receiving allergen immunotherapy injections, *J. Allergy Clin. Immunol.* 86 (3 Pt 1) (1990 Sep) 407–411.
- [16] L.P. Chung, G. Waterer, P.J. Thompson, Pharmacogenetics of beta2 adrenergic receptor gene polymorphisms, long-acting beta-agonists and asthma, *Clin. Exp. Allergy* 41 (3) (2011 Mar) 312–326 [Review].
- [17] R.F. Lemanske Jr., D.T. Mager, C.A. Sorkness, D.J. Jackson, S.J. Boehmer, F.D. Martinez, et al., Step-up therapy for children with uncontrolled asthma receiving inhaled corticosteroids, *N. Engl. J. Med.* 362 (11) (Mar 18 2010) 975–985 [Multicenter Study Randomized Controlled Trial Research Support, N.I.H., Extramural].
- [18] S.P. Peters, S.J. Kunselman, N. Icitovic, W.C. Moore, R. Pascual, B.T. Ameredes, et al., Tiotropium bromide step-up therapy for adults with uncontrolled asthma, *N. Engl. J. Med.* 363 (18) (Oct 28 2010) 1715–1726 [Comparative Study Multicenter Study Randomized Controlled Trial Research Support, N.I.H., Extramural].
- [19] R.J. Martin, S.J. Szefer, T.S. King, M. Kraft, H.A. Boushey, V.M. Chinchilli, et al., The Predicting Response to Inhaled Corticosteroid Efficacy (PRICE) trial, *J. Allergy Clin. Immunol.* 119 (1) (Jan 2007) 73–80 [Multicenter Study Randomized Controlled Trial Research Support, N.I.H., Extramural].

- [20] B.K. Kobilka, T. Friele, H.G. Dohlman, M.A. Bolanowski, R.A. Dixon, P. Keller, et al., Delineation of the intronless nature of the genes for the human and hamster beta 2-adrenergic receptor and their putative promoter regions, *J. Biol. Chem.* 262 (15) (May 25 1987) 7321–7327 [Comparative Study].
- [21] G.A. Hawkins, K. Tantisira, D.A. Meyers, E.J. Ampleford, W.C. Moore, B. Klanderman, et al., Sequence, haplotype, and association analysis of ADRbeta2 in a multiethnic asthma case–control study, *Am. J. Respir. Crit. Care Med.* 174 (10) (Nov 15 2006) 1101–1109 [Research Support, N.I.H., Extramural].
- [22] S.A. Green, G. Cole, M. Jacinto, M. Innis, S.B. Liggett, A polymorphism of the human beta 2-adrenergic receptor within the fourth transmembrane domain alters ligand binding and functional properties of the receptor, *J. Biol. Chem.* 268 (31) (Nov 5 1993) 23116–23121 [Research Support, U.S. Gov't, P.H.S.].
- [23] S.A. Green, J. Turki, I.P. Hall, S.B. Liggett, Implications of genetic variability of human beta 2-adrenergic receptor structure, *Pulm. Pharmacol.* 8 (1) (Feb 1995) 1–10 [Research Support, U.S. Gov't, P.H.S. Review].
- [24] S.A. Green, J. Turki, M. Innis, S.B. Liggett, Amino-terminal polymorphisms of the human beta 2-adrenergic receptor impart distinct agonist-promoted regulatory properties, *Biochemistry* 33 (32) (Aug 16 1994) 9414–9419 [Research Support, U.S. Gov't, P.H.S.].
- [25] J. Grainger, K. Woodman, N. Pearce, J. Crane, C. Burgess, A. Keane, et al., Prescribed fenoterol and death from asthma in New Zealand, 1981–7: a further case–control study, *Thorax* 46 (2) (Feb 1991) 105–111 [Research Support, Non-U.S. Gov't].
- [26] N. Pearce, The use of beta agonists and the risk of death and near death from asthma, *J. Clin. Epidemiol.* 62 (6) (Jun 2009) 582–587 [Historical Article Research Support, Non-U.S. Gov't].
- [27] N. Pearce, C. Burgess, J. Crane, R. Beasley, Fenoterol, asthma deaths, and asthma severity, *Chest* 112 (4) (Oct 1997) 1148–1150 [Comment Letter].
- [28] K. Basu, C.N. Palmer, R. Tavendale, B.J. Lipworth, S. Mukhopadhyay, Adrenergic beta(2)-receptor genotype predisposes to exacerbations in steroid-treated asthmatic patients taking frequent albuterol or salmeterol, *J. Allergy Clin. Immunol.* 124 (6) (Dec 2009) 1188–94 [Research Support, Non-U.S. Gov't].
- [29] M.E. Wechsler, S.J. Kunselman, V.M. Chinchilli, E. Bleecker, H.A. Boushey, W.J. Calhoun, et al., Effect of beta2-adrenergic receptor polymorphism on response to longacting beta2 agonist in asthma (LARGE trial): a genotype-stratified, randomised, placebo-controlled, crossover trial, *Lancet* 374 (9703) (Nov 21 2009) 1754–1764 [Multicenter Study Randomized Controlled Trial Research Support, N.I.H., Extramural].
- [30] A. de Alarcon, J.W. Steinke, R. Caughey, E. Barekzi, K. Hise, C.W. Gross, et al., Expression of leukotriene C4 synthase and plasminogen activator inhibitor 1 gene promoter polymorphisms in sinusitis, *Am. J. Rhinol.* 20 (5) (Sep-Oct 2006) 545–549 [Comparative Study].
- [31] D.D. Stevenson, B.L. Zuraw, Pathogenesis of aspirin-exacerbated respiratory disease, *Clin. Rev. Allergy Immunol.* 24 (2) (Apr 2003) 169–188 [Review].
- [32] X.S. Wang, A.Y. Wu, P.S. Leung, H.Y. Lau, PGE suppresses excessive anti-IgE induced cysteinyl leucotrienes production in mast cells of patients with aspirin exacerbated respiratory disease, *Allergy* 62 (6) (Jun 2007) 620–627 [Research Support, Non-U.S. Gov't].
- [33] M.P. Berges-Gimeno, R.A. Simon, D.D. Stevenson, The effect of leukotriene-modifier drugs on aspirin-induced asthma and rhinitis reactions, *Clin. Exp. Allergy* 32 (10) (Oct 2002) 1491–1496 [Research Support, Non-U.S. Gov't Research Support, U.S. Gov't, P.H.S.].
- [34] M.P. Berges-Gimeno, R.A. Simon, D.D. Stevenson, Long-term treatment with aspirin desensitization in asthmatic patients with aspirin-exacerbated respiratory disease, *J. Allergy Clin. Immunol.* 111 (1) (Jan 2003) 180–186 [Research Support, Non-U.S. Gov't Research Support, U.S. Gov't, P.H.S.].
- [35] H. Rizk, Role of aspirin desensitization in the management of chronic rhinosinusitis, *Curr. Opin. Otolaryngol. Head Neck Surg.* 19 (3) (Jun 2011) 210–217.
- [36] D.D. Stevenson, Aspirin desensitization in patients with AERD, *Clin. Rev. Allergy Immunol.* 24 (2) (Apr 2003) 159–168 [Review].
- [37] A.N. Williams, K.M. Woessner, The clinical effectiveness of aspirin desensitization in chronic rhinosinusitis, *Curr. Allergy Asthma Rep.* 8 (3) (May 2008) 245–252 [Review].
- [38] L.J. Collins, D. Penny, The RNA infrastructure: dark matter of the eukaryotic cell? *Trends Genet.* 25 (3) (Mar 2009) 120–128 [Research Support, Non-U.S. Gov't Review].
- [39] M.C. Weinstein, J.A. Skinner, Comparative effectiveness and health care spending –implications for reform, *N. Engl. J. Med.* 362 (5) (Feb 4 2010) 460–465 [Research Support, N.I.H., Extramural Research Support, Non-U.S. Gov't].
- [40] A.M. Garber, S.R. Tunis, Does comparative-effectiveness research threaten personalized medicine? *N. Engl. J. Med.* 360 (19) (May 7 2009) 1925–1927.