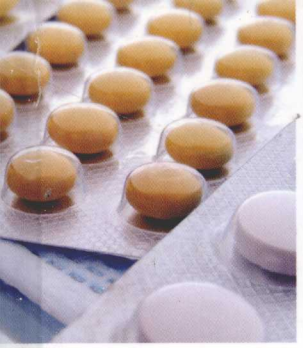

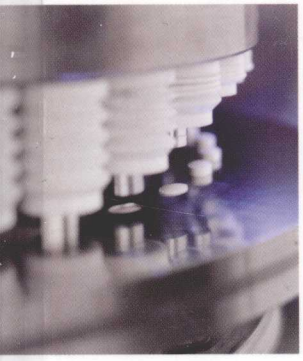

Genomic Biomarkers *for* Pharmaceutical Development

Advancing Personalized Health Care

Edited by

Yihong Yao
Bahija Jallal
Koustubh Ranade

GENOMIC BIOMARKERS FOR PHARMACEUTICAL DEVELOPMENT

ADVANCING PERSONALIZED HEALTH CARE

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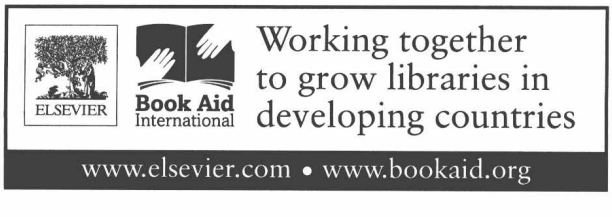


GENOMIC BIOMARKERS FOR PHARMACEUTICAL DEVELOPMENT

Preface

A number of books have been published lately on genomics and personalized medicine. We felt, however, that none was comprehensive and reflected adequately our thinking and lived experience in applying genomics to drug development and personalized healthcare. Furthermore, published literature was also relatively silent on the translational science approaches to identify the right therapeutic for the right patient that we believe will be needed to improve the probability of success of clinical trials, particularly in the early stages of drug development.

We made an effort to address these gaps in this book by describing genomic strategies to explore basic questions in drug development: What kind of patient might most benefit from a new therapeutic? Which patient might experience adverse effects? How does one identify such patients? How does one determine if a new therapeutic is having the desired effect even before clinical symptoms improve? The most current thinking on these questions from industry, academia and the government is assembled in this book.

The book begins with a broad discussion of how to employ genomics to develop translational approaches for early stage clinical trials and to understand adverse side effects of therapeutics. We include case studies to illustrate key points. The ensuing chapters describe in detail how strategies outlined in the first chapter may be reduced to practice in different therapeutic areas.

Given the vast investment in the war on cancer over the past 40 years, and the breathtaking progress in understanding the complex landscape of the cancer genome, it should come as no surprise that personalized healthcare based on genomics is most mature for this therapeutic area. As the chapter on cancer makes clear, however, even in this area, significant hurdles need to be overcome before individualized therapies come into routine clinical practice.

The chapter on autoimmune diseases describes early steps in developing potential personalized healthcare for rheumatoid arthritis and systemic lupus erythematosus, and the use of genomic biomarkers to determine if a therapeutic is having the intended effect. Chapter 4 is a tour de force of how to apply genomics to tease apart the complexity of asthma and turn that knowledge into a targeted therapeutic. The nascent fields of microRNAs – small RNA molecules that regulate the expression of a large number of genes – and toxicogenomics are the subjects of the next two chapters. The book ends with a discussion of the regulatory process for obtaining approval for diagnostic tests that are necessary for personalized healthcare. As our understanding of cardiovascular diseases, mental illness and rare genetic disorders deepens, we look forward to discussing targeted therapeutics for these diseases in a future editions of this book.

The book is intended for anyone interested in understanding current and emerging practices in translational science as they

relate to genomics and drug development. It is appropriate for advanced graduate students, clinicians and scientists working in the bio-pharmaceutical industry.

We are very grateful to the co-authors for their efforts and thank our families for tolerating the many evenings and weekends that were devoted to this book.

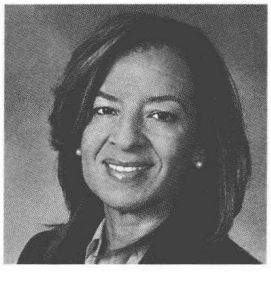
Yihong Yao
Bahija Jallal
Koustubh Ranade

About the Editors

Dr. Yihong Yao is director and head of Pharmacogenomics and Bioinformatics group at MedImmune, LLC. The focus of Dr. Yao's group is the utilization of cutting edge genomic and genetic approaches to developing pharmacodynamic and predictive diagnostic markers to understand disease linkage and to identify the right patients that might respond (or not respond) to therapeutic interventions. The other areas of interest in Dr. Yao's group include: unveiling potential key drivers in cancer, and in respiratory and inflammatory diseases, and understanding the role of miRNAs in disease pathogenesis.

Dr. Yao received a Bachelor's degree in Biochemistry from Fu Dan University in 1988. He received a Master's degree in Bioinformatics from Boston University. He completed his doctorate in Biochemistry and Biophysics from the University of Kansas in Lawrence, Kansas. He conducted his postdoctoral research at Johns Hopkins Medical School in Baltimore, Maryland.

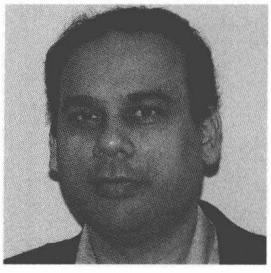
Dr. Yao has authored over 50 peer-reviewed publications, edited a book and has over 15 patents.



Dr. Bahija Jallal is Executive Vice President of AstraZeneca and head of MedImmune, a global biologics research and development organization with locations in Gaithersburg, California and Cambridge, UK. She is a member of the senior executive team at AstraZeneca reporting to the CEO. Dr. Jallal joined MedImmune in March 2006. She has guided the MedImmune R&D organization through the unprecedented growth and expansion of its biologics pipeline from 40 drugs to more than 120 today. She inspires creativity, out-of-the-box thinking and a dedication to scientific excellence in the more than 2,500 employees at MedImmune.

Dr. Jallal received a Master's degree in biology from the Universite de Paris VII in France, and her doctorate in physiology from the University of Pierre & Marie Curie in Paris. She conducted her postdoctoral research at the Max-Planck Institute of Biochemistry in Martinsried, Germany.

Dr. Jallal has authored over 70 peer-reviewed publications and has over 15 patents. She is a member of the American Association of Cancer Research, the American Association of Science and the Pharmacogenomics Working Group. She serves as a member of the Board of Directors for the Association of Women in Science and is an advisory member of the Healthcare Business Women's Association. She was named one of FierceBiotech's 2011 Women in Biotech and in 2012, she received Washington Business Journal's Women Who Mean Business Award.



Dr. Koustubh Ranade is Fellow in Translational Sciences at MedImmune, where he is responsible for leading translational and personalized healthcare strategies for multiple drug development programs across therapeutic areas. Prior to MedImmune, Koustubh was Associate Group Medical Director of Clinical Diagnostics at Genentech and Director of Human Genetics at Bristol-Myers Squibb. Dr Ranade has published extensively on human genetics and pharmacogenomics and is inventor on four issued patents. He received his PhD in Molecular Genetics from the University of Massachusetts Medical School, and did his postdoctoral training at the National Human Genome Research Institute and Stanford University.

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CHAPTER

1

Application of Translational Science to Clinical Development

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1.1 INTRODUCTION

The pharmaceutical industry is in crisis. In the year 2012 alone, branded drugs valued at over $30 billion lost patent protection, thereby allowing generic manufacturers to make and sell lower-priced versions of blockbuster drugs [1]. The industry as a whole has been unsuccessful in replacing drugs going off-patent with sufficient new molecular entities (NMEs). Despite staggering investment in R&D – the top ten pharmas spent $60 billion on R&D in 2010 – the number of approvals has changed little over the past decade (Fig. 1.1).

This level of investment without commensurate improvement in approvals of new medicines is likely unsustainable and has, in fact, contributed to waves of mergers in the pharmaceutical industry accompanied by tens of thousands of layoffs in 2007–2012.

Many reasons have been attributed to the lack of apparent productivity in big pharma, but, as the graph in Fig. 1.2 indicates, it is likely that the main culprit is the low probability of success (PoS), perhaps as low as 10%, of investigational drugs entering Phase II clinical trials that are eventually approved [3]. Coupled with the high cost of clinical trials, this low PoS makes drug development a highly risky proposition, and drives the industry to invest in already validated targets that are more likely to yield approvable, albeit less innovative, drugs, at the end of a multi-year effort.

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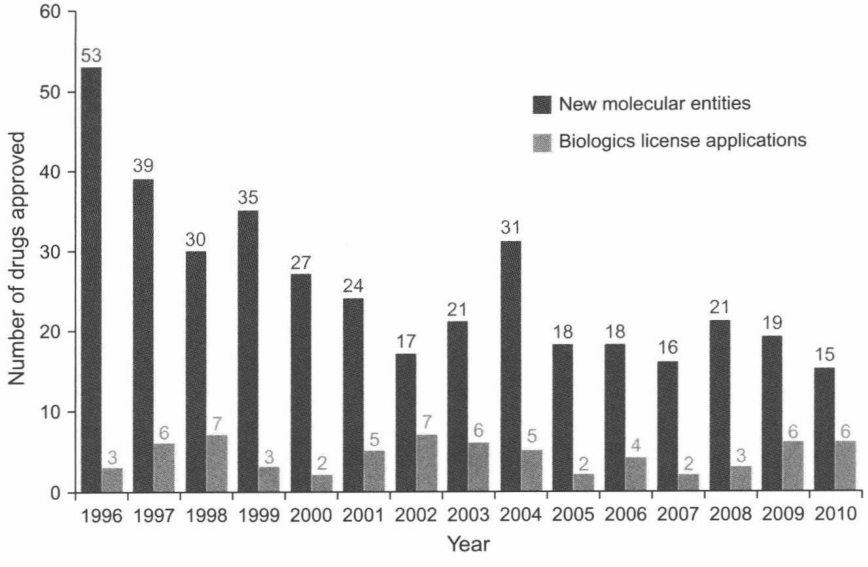

FIGURE 1.1 Number of drug approvals, small molecules and biologics. *From [2].*

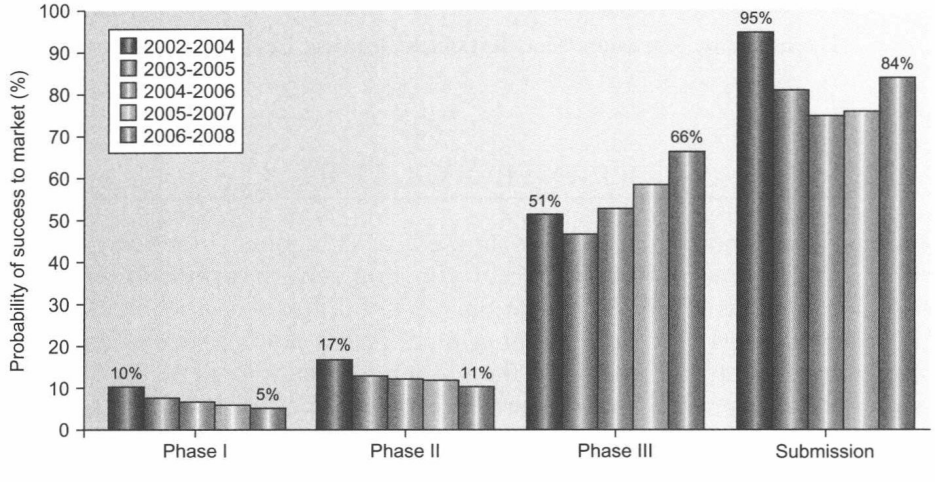

FIGURE 1.2 Probability of success to market from key clinical development milestones. *From [3].*

Paradoxically, it seems that there has never been a better time in biomedical research with all the innovations we are currently witnessing. The Human Genome Project and ancillary efforts, the development of next-generation sequencing and other large scale genome analysis tools have transformed our understanding of diseases, especially cancer [4]. Not a day goes by without news of identification of a 'gene for' a common disease or new insight into a pathway that drives a common cancer. One of the key challenges for pharma R&D will be to translate this explosion in genomic knowledge and new insights into disease pathways

into innovative therapeutics that extend and enhance the lives of patients with unmet medical needs.

We believe that judicious application of genomic analysis to develop greater understanding of the molecular underpinnings of complex diseases such as cancer, rheumatic and respiratory diseases will lead to novel targets and therapeutics that are tailored to subsets of these diseases. Together with biomarkers that identify subsets of patients likely to benefit (or not) from targeted therapies, such therapeutics are likely to have a greater PoS in clinical development than those that target all-comers. In this chapter we describe current and emerging translational strategies to apply our expanding genomic knowledge to this end. For the purpose of this discussion, we define 'translational science' as treating the 'right' patient with the 'right drug' at the 'right dose'. Our objective here is to illustrate broad strategies for identifying the right patient, the right drug and right dose using examples from the literature. We include in our definition of the right patient not only those that will benefit from a novel therapeutic but also those that are less likely to be harmed by it, and we end the chapter with a discussion about adverse drug reactions.

1.2 TWO APPROACHES TO IDENTIFY PATIENT SUBSETS THAT ARE LIKELY TO RESPOND TO INDIVIDUAL THERAPEUTIC INTERVENTIONS

The current paradigm for drug development is to test a new drug candidate in a variety of diseases, such as different types of solid tumors including prostate, breast, colon or hematologic malignancies. Depending on whether a positive signal in a small trial is observed, a couple of indications are selected to conduct follow up larger registrational trials. While this approach has been successful in the past, it is also a key contributor to the ever-increasing cost of drug development; perhaps more importantly it exposes many patients to therapies from which they are unlikely to benefit because the disease of interest is not primarily driven by the targeted pathway in all patients. Viewed from a slightly different perspective, this approach in effect uses the investigational drug to probe the underlying disease in a given patient to assess whether it is amenable to the pathway that is targeted by the drug.

An emerging approach, which is outlined in Fig. 1.3, is to understand, using genomic approaches (e.g., sequencing of tumors or gene expression analysis of relevant tissue), heterogeneity of disease first and thus identify subsets of patients in whom a biological pathway is activated by mutation or by elevated expression of genes in the pathway. The disease in such patients may, therefore, be causatively linked to a particular biological pathway and thus be amenable to therapeutics targeted to the pathway.

Such patients – the right patients – are then targeted with a therapeutic that is tailored to them – the right drug. Key to the success of this approach is a reliable way to identify such patient subsets, i.e., a companion diagnostic to the tailored therapeutic that is economically viable and can be easily implemented in the clinic. We illustrate this approach using the example of crizotinib (Xalkori®) from Pfizer, an ALK (anaplastic lymphoma kinase) and ROS1 (c-ros oncogene1, receptor tyrosine kinase) inhibitor that was approved in 2011 for patients with a particular kind of non-small cell lung cancer (NSCLC) [6].

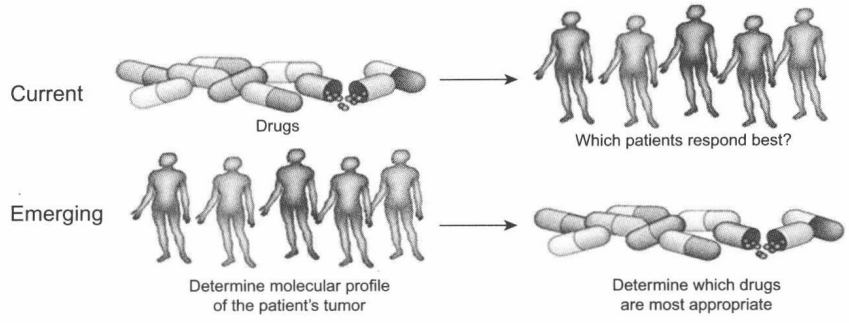

FIGURE 1.3 Two approaches to drug development: In the past, new molecular entities were tested in a variety of indications, e.g., cancers of different types, to identify those patients most likely to respond. In the emerging translational approach, molecular heterogeneity of a disease is analyzed first, and then therapeutics are developed and tailored to subsets of disease. *Adapted from [5].*

1.2.1 Prospective Analysis: The Case of Crizotinib in NSCLC

The crizotinib story started several years ago, when analysis of a cDNA library from a Japanese patient with lung adenocarcinoma identified a novel fusion between the EML4 and ALK genes with the ability to transform 3T3 fibroblasts [7]. Analysis of a series of biopsies from NSCLC patients revealed that ~5% of patients carry this fusion protein.

Soon after the publication of the initial discovery in 2007, it was found that crizotinib, a small molecule inhibitor of the protein encoded by the ALK gene, was very effective in NSCLC patients whose tumors harbored the ALK fusion gene. It caused tumors to shrink or stabilize in 90% of 82 patients carrying the ALK fusion gene, and tumors shrank at least 30% in 57% of people treated [8]. These promising clinical results led to a Phase II and a Phase III trial, which selectively enrolled NSCLC patients with ALK fusion genes. Astonishingly, within four years of the initial publication by Soda et al., the Food and Drug Administration (FDA) approved crizotinib for the treatment of certain late stage (locally advanced or metastatic) NSCLC patients whose tumors have ALK fusion genes as identified by a companion diagnostic that was approved simultaneously with the drug [6].

There are several important lessons to be learned from the development of crizotinib. First, understanding molecular heterogeneity to identify a mutation or pathway that is causally linked to the disease is crucial to the eventual success. With this knowledge in hand, investigators could design small but highly effective trials targeted to those patients more likely to benefit from the therapy. Such approaches allow drug companies to save both money and time in drug development. The approval for crizotinib was based on two registrational trials that enrolled fewer than 150 subjects each. To better illustrate how targeting patients can improve the PoS of a clinical trial, we performed simulations to estimate the sample sizes that would be required if patients had not been selected in trials of a drug like crizotinib that is targeted to, for instance, only 10% of the population.

Under the assumption of placebo response rates ranging from 6–14% in typical cancer clinical trials [9], if patient randomization is conducted requiring the presence of a biomarker, or biomarker positive group, the minimum sample size needed at 80% power and alpha = 0.05 could be as low as N = 33/arm, with a 30% effect size and 6% response rate in

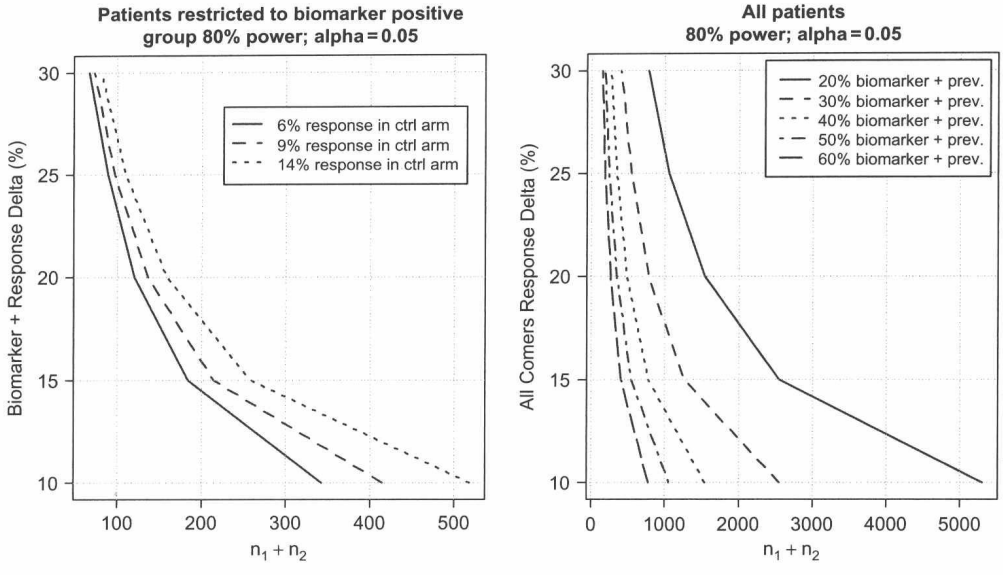

FIGURE 1.4 (left) Relationship between effect size and total sample size when restricting patient inclusion to biomarker positive patients under different control arm response rates (80% power and alpha = 0.05), and (right) the same association showing different levels of biomarker positive patient prevalence, assuming 6% control arm response rate with no restriction to biomarker positive patients, and only the biomarker positive patients showing improvement (80% power and alpha = 0.05). Note that total response delta is plotted on the y axis (right), though sample sizes are calculated using the reduced response delta as explained in the text.

the control arm to as high as N = 259/arm with a 10% effect size and 14% response rate in the control arm [10] (Fig. 1.4 left).

In contrast to this trial design where only biomarker positive patients are included, the sample size requirement in an all-comers trial design, i.e., without selectively enrolling patients, is driven not only by effect size, but by the prevalence of patients with the particular disease sub-type (e.g., NSCLC patients with ALK fusion). For example, assuming a 6% response rate in the control arm, if the trial is not restricted to biomarker positive patients (i.e., those with ALK fusion in this example), and we assume the same effect size in the previous example of 30%, if 10% of the patients are identified as biomarker positive (and only these patients show improvement), the overall improvement rate would be reduced to 3%. Under this design, 1274 patients/arm would be required at alpha = 0.05 and 80% power. If the biomarker positive patient prevalence is identified to be 30%, the reduced effect size is 9% and expected sample size is reduced to 202/arm, under the same assumptions (Fig. 1.4 right). This example illustrates how easily sample size requirements can be affected by either reduced biomarker positive patient prevalence or decreased effect sizes in a clinical trial.

The second lesson from the crizotinib example is the importance of developing strong testable hypotheses early. Although developing robust and reliable hypotheses is often easier said than done, with the right approach equipped with the powerful technologies we currently have, such hypotheses are not out of reach. Fortuitously, in the case of crizotinib one of the clinical sites in enrolling patients in the Phase I trial was already developing tools

to assess ALK fusion genes and was able to quickly translate published results into clinical development.

The foregoing discussion has focused on cancer, but similar prospective approaches have been applied to inflammatory diseases as well. As Arron and Harris describe in their chapter on asthma (Chapter 4), gene expression analysis of lung epithelial tissue from treatment-naïve asthma patients revealed that a subset of patients had significantly elevated expression of genes that were regulated by the cytokine IL13. After substantial follow up, a clinical trial demonstrated that this subset of patients, which could be identified with a serum biomarker, derived significant benefit from a novel anti-IL13 therapeutic. This initial observation needs to be confirmed in ongoing Phase III trials, but demonstrates the power of this translational approach. An analogous translational approach to identify a subset of patients with systemic lupus erythematosus is described in the chapter on autoimmune diseases (Chapter 3).

1.2.1.1 Retrospective Analysis to Identify Responders

In contrast to the prospective approaches described above to identify patients who may derive benefit from a therapeutic, we describe below successful examples to identify predictive markers by comparing responders and non-responders, i.e., from retrospective analysis of clinical trials.

1.2.1.2 Large Molecule Inhibitors of Epidermal Growth Factor Receptor (EGFR)

The monoclonal antibodies cetuximab and panitumumab which are targeted against the EGFR have been approved for the treatment of metastatic colorectal cancer. Initial analysis of a small number of responders and non-responders for mutations in genes in the EGFR signaling pathway – KRAS, BRAF, PI3KCA – revealed that KRAS mutations were readily detected in non-responders but not in responders [11]. Of the 11 patients who responded to cetuximab, none was mutant for KRAS; in contrast 13 of 19 non-responders were KRAS mutants. These significant results were confirmed in subsequent large trials of cetuximab [12] and panitumumab [13]. Although the FDA guidance calls for prospective stratification of clinical trials to provide an adequate test of a predictive marker (see Chapter 7), in this case, KRAS mutation status as a predictor of response was approved as a companion diagnostic for cetuximab and panitumumab because of overwhelming evidence from multiple retrospective analyses. Further details of KRAS and response to EGFR targeted therapies can be found in Chapter 2.

1.2.1.3 Small Molecule Inhibitors of EGFR: Case of Gefitinib

The small molecule inhibitors of EGFR, gefitinib (IRESSA™, AstraZeneca) and erlotinib (Tarceva®, Roche) were initially approved in all-comers based on standard registrational trials [14,15]. It was several years post-approval that it was discovered that these inhibitors provide significant benefit to NSCLC patients carrying a particular tumor biomarker, a mutation within the EGFR gene [16–18]. Encouraging anti-tumor activity was observed in NSCLC in a Phase I trial [19]. In two subsequent Phase II trials (IDEAL 1 and IDEAL 2), promising and well-tolerated drug activity was observed. In the same trials, EGFR protein expression levels within the tumor were tested as a potential predictive biomarker for clinical response, but no relationship was found between EGFR protein expression and response [20,21].