1 Definition: Translational and Personalised Medicine, Biomarkers, Pharmacodynamics

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Introduction

Our understanding of the complex underlying mechanisms of cancer development and progression has improved with the application of novel laboratory-based techniques including DNA sequencing, gene expression profiling, analysis of DNA methylation, and proteomics. Following the publication of the first full-length human genome sequence in 2001, there have been many large-scale sequencing initiatives such as the Cancer Genome Atlas (TCGA; http://cancergenome.nih.gov) and the International Cancer Genome Consortium (ICGC; https://icgc.org), projects that have characterised genomic alterations in solid tumours. These investigations have uncovered key mutations and molecular pathways involved in oncogenesis and cell proliferation that have an important impact on the diagnosis, prognosis, and treatment of patients and have enabled the tailoring of anti-cancer therapy in clinical practice. These discoveries also have important implications for individuals at high risk for developing certain solid tumours by identifying patients who may benefit from more intensive screening. For example, women with pathogenic germline mutations in the *BRCA1* or *BRCA2* tumour suppressor genes, involved in DNA repair of double-stranded breaks, are predisposed to developing breast, ovarian, and other cancers and are candidates for more specialised cancer screening programmes.

In addition to advances in molecular biology techniques, there have also been developments in diagnostic imaging and histopathology that have enabled more accurate diagnosis and characterisation of solid tumours and have provided for better monitoring of tumour response to treatment. For example, the development of fluorescence in situ hybridisation (FISH) techniques for use in the clinical laboratory has facilitated the routine testing of breast tumours for *ERBB2* (*HER2/neu*) gene amplification in cases where immunohistochemistry staining is equivocal.

The identification of common genetic alterations in solid tumours, along with the development of high-throughput and cost-effective molecular diagnostics, is paving the way to enable the individualisation of cancer treatment.

Defining Translational Research and Personalised Medicine

The application of discoveries and technologies from the basic science research setting to the clinical setting is the basis for translational research that has facilitated the identification of novel drug targets and treatment strategies. This has ushered in a new era of a more individualised, or personalised, approach to cancer treatment, particularly in the metastatic setting. The standard of care for many patients with advanced malignancies is gradually evolving from empirical treatment based on clinical–pathological characteristics to the use of targeted approaches based on the molecular profile of the tumour. Fundamental insights into cancer biology gained from preclinical studies are used to design human clinical trials to test novel approaches to diagnosis or therapy. Results from phase III studies are then ultimately incorporated into everyday clinical practice. In designing clinical trials with novel targeted agents, patient selection is important, as agents targeted toward a particular genetic alteration are often inactive and even harmful in an unselected patient population. For example, initial phase III studies of epidermal growth factor receptor (EGFR) inhibitors in patients with metastatic colorectal cancer did not show a benefit for their use in an unselected population, but showed a survival benefit in patients with wild-type *KRAS*. In melanoma, the discovery of genetic alterations that drive tumour progression has led to a number of targeted therapies including drugs that target the *BRAF* V600E

mutation. In patients with this mutation, treatment with the BRAF inhibitor vemurafenib induced tumour regression and led to improved overall survival when compared to chemotherapy.

Translational research in oncology has been greatly facilitated by rapid advances in molecular biology and histopathology techniques. For example, new methods for tumour acquisition and histopathology analysis can identify subpopulations of cells with unique mechanisms of sensitivity or resistance to specific therapies. Such interpatient and intrapatient tumour heterogeneity can influence prognosis and responses to systemic therapies. The term "personalised medicine" refers to the application of patient-specific genetic information (both germline and somatic) and molecular and/or cellular tumour characteristics to select the optimal treatment for individual patients with the goal of improved therapeutic efficacy and reduced toxicity. It involves the use of biomarkers that provide unique patient- and tumour-specific molecular information.

Biomarkers are molecular or cellular characteristics that indicate a normal or pathogenic process that can be used to aid in diagnosis, defining susceptibility for a particular disease, and determining clinical outcomes or response to a specific therapy or intervention. They can also be used to facilitate the development of rational drug combinations and to identify potential resistance mechanisms.

In addition, biomarkers provide clues about the mechanism of action of a particular drug and serve as a tool for selecting the most appropriate patients for enrolment in early phase clinical trials based on molecular characteristics of the tumour. The discovery of specific mutations that predict the efficacy of a particular drug in a molecularly defined patient cohort has greatly transformed drug development programmes in oncology, shifting the focus from the development of non-specific cytotoxic chemotherapies to molecularly targeted therapeutics.

Prognostic Biomarkers

A prognostic marker is measured before treatment and identifies tumour-specific molecular or histopathological characteristics including somatic or germline mutations, changes in DNA methylation, micro-RNA levels,

or circulating tumour cells in blood that are associated with long-term outcome or course of a disease. As shown in Figure 1, for a prognostic biomarker, the probability of survival is related to whether or not the biomarker is expressed. In patients who express the biomarker of interest, survival is similar between treated and untreated patients.

Prognostic biomarkers allow for the selection of patients who need more intensive surveillance or adjuvant therapy. In acute myeloid leukaemia (AML), cytogenetic abnormalities serve as prognostic biomarkers for risk categorisation. Inversions in chromosome 16 as well as translocations between chromosomes 8 and 21 and chromosomes 15 and 17 are associated with a favourable prognosis, while deletions in chromosomes 5 and 7 are associated with an unfavourable prognosis. In multiple myeloma, levels of beta-2 microglobulin and albumin are used as prognostic markers to stage myeloma and to classify patients into favourable, intermediate, or unfavourable overall survival prognoses prior to initiating systemic therapy. In breast cancer, commonly used prognostic markers include tumour size, nodal status, grade, and presence or absence of lymphovascular invasion.

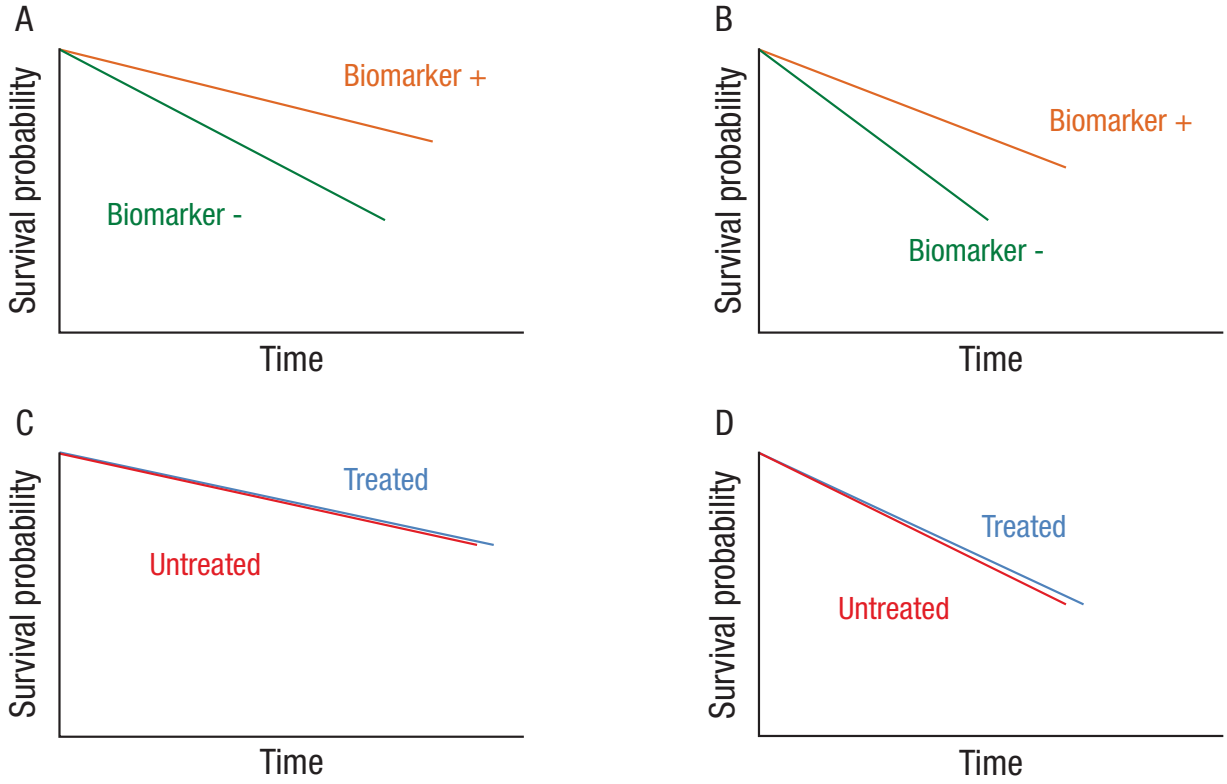

Figure 1 *Prognostic biomarkers: Treated (A) and untreated patients (placebo or best supportive care) (B). Patients expressing (C) and not expressing (D) the biomarker of interest.*

Predictive Biomarkers

A predictive biomarker is usually measured before treatment and provides information on the probability of response to a particular therapy. As depicted graphically in Figure 2, the probability of survival depends on treatment in those patients who express the biomarker associated with response to a particular therapy. For patients who are biomarker negative, there is no difference in survival between treated and untreated patients. An example of a predictive biomarker in breast cancer is expression of the HER2/neu protein. Approximately 15% to 20% of patients with invasive breast cancer have increased expression of the HER2/neu protein (a member of the EGFR family of transmembrane receptors) that is associated with response to anti-HER2-targeted agents such as trastuzumab, pertuzumab, and trastuzumab–emtansine. Another example is expression of the oestrogen receptor in breast cancer, which correlates with sensitivity to hormonal agents used in the adjuvant and metastatic settings.

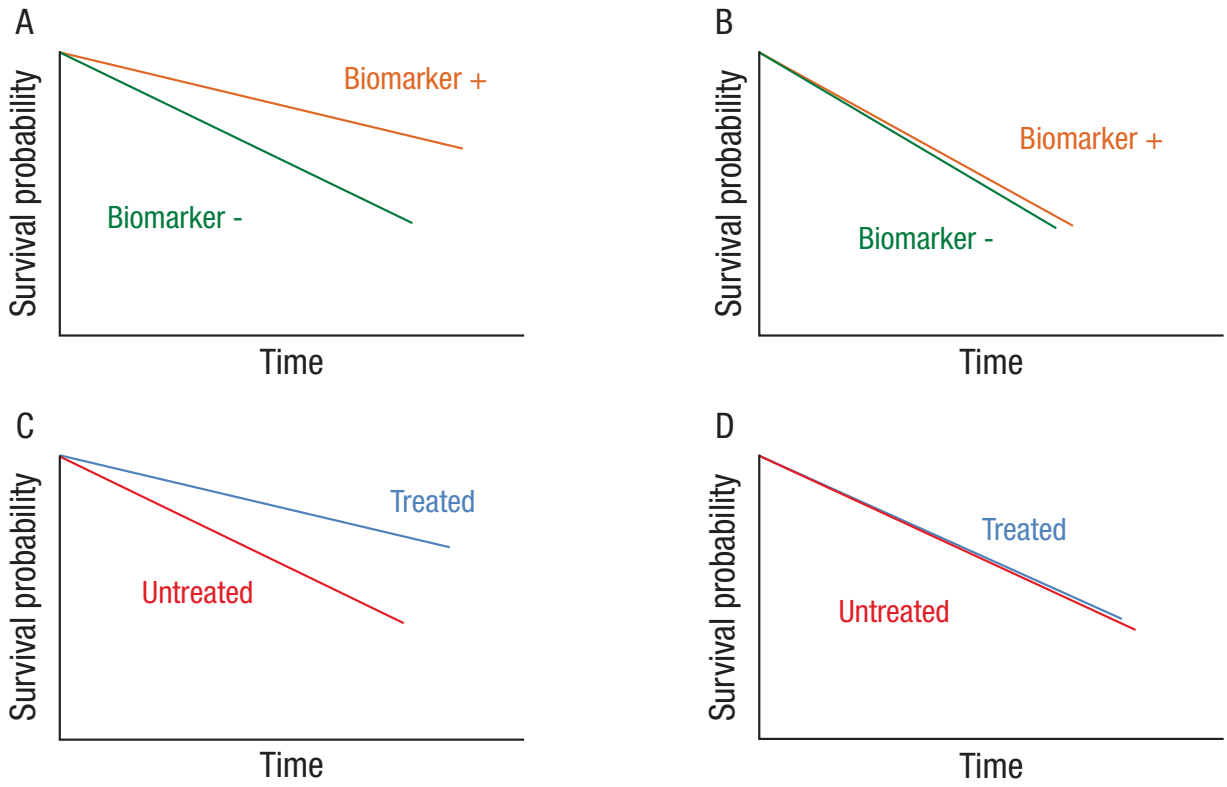

Figure 2 *Predictive biomarkers: Treated (A) and untreated patients (placebo or best supportive care) (B). Patients expressing (C) and not expressing (D) the biomarker of interest.*

In metastatic non-small cell lung cancer, patients with tumours harbouring either *EGFR* exon 19 or 21 gene mutations or the *EML4-ALK* fusion protein can be offered oral targeted therapies against EGFR (gefitinib or erlotinib) or ALK (crizotinib), respectively, which are more effective than cytotoxic chemotherapy. These somatic genetic alterations are only identified in a subset of metastatic non-small cell lung cancer patients (*EGFR* mutations in 15%-20% and *EML4-ALK* translocation in 3%-5%). The development of robust, clinically applicable biomarkers identifying these alterations has enabled the selection of only those patients who would derive benefit from targeted therapy.

In colorectal cancer, *KRAS* is a frequently mutated oncogene and is a predictive biomarker for resistance to anti-EGFR monoclonal antibody therapy. Approximately 40% of colorectal cancers have mutations in *KRAS*. This is routinely tested in the metastatic setting, as only tumours with wild-type *KRAS* derive benefit from EGFR inhibitors such as cetuximab and panitumumab. More recently, it has been shown that additional mutations in other *RAS* genes are observed in 5%-10% of colorectal cancers and are also associated with resistance to anti-EGFR therapy. *KRAS* mutations were initially identified retrospectively after large clinical trials were completed in unselected patients. This example highlights the importance of tumour tissue collections in larger clinical trials in order to facilitate the identification of clinically useful biomarkers in the future.

Biomarker Selection and Validation

Most prognostic biomarkers are not routinely used in clinical practice, as they are generally developed in unfocused clinical studies composed of samples of heterogeneous patients with available tissues. These studies, which are not specifically designed to address the clinical significance of a prognostic biomarker, do not result in validated biomarkers with clinical utility. In order for a biomarker to be clinically useful, it should correlate with tumour behaviour and/or treatment outcomes. Its use in guiding treatment decisions should lead to improved clinical outcomes. Since a particular biomarker may be useful in one disease but not in another, it is also necessary to define the disease in which it should be used. Selected biomarkers used in routine clinical decision-making for advanced solid tumours are listed in Table 1.

Table 1 *Selected Biomarker Tests Routinely Used in Clinical Decision-making for Advanced Solid Tumours. Modified from Bedard PL, Hansen AR, Ratain MJ, et al. Tumor heterogeneity in the clinic. Nature 2013; 501:355–364.*

Tumour type	Biomarker	Prognostic or predictive biomarker
Oligodendroglioma	1p and 19q co-deletion *MGMT* promoter methylation	Prognostic/predictive Prognostic/predictive
Medullary thyroid	*RET* mutation	Prognostic
Breast	ER expression PR expression *HER2* amplification	Prognostic/predictive Prognostic Prognostic/predictive
Lung	*EGFR* mutation *EML4-ALK* translocation	Prognostic/predictive Prognostic/predictive
Gastric	*HER2* amplification	Prognostic/predictive
Colorectal	*KRAS* mutation	Predictive
Melanoma	*BRAF* mutation	Prognostic/predictive
Gastrointestinal stromal	*KIT* mutation *PDGFRA* mutation	Predictive Predictive

ER, oestrogen receptor; PR, progesterone receptor

In order to be clinically useful, a biomarker should be assayed in a specimen that is easily accessible in the clinical setting and can be readily obtained with standardised collection and processing protocols. Biomarkers that require serial assessments over a long period of time should be able to be collected in a minimally-invasive manner to be clinically feasible. The biomarker assay itself should be specific to the disease type tested and be reproducible, with appropriate cutoffs indicating the presence or absence of the biomarker. It is important for biomarkers to be validated in an independent clinical population that differs from the population used to develop the biomarker. Use of a particular biomarker should lead to a clinical decision that is linked to clinically meaningful outcomes, such as improvement in survival or quality of life or a decreased toxicity.

Pharmacodynamics and Drug Development

The incorporation of pharmacodynamic biomarkers in early phase drug development can provide important information about the biological

effects of the treatment intervention on the patient and tumour. In oncology, "pharmacodynamics" refers to the effect of the drug on the patient and the tumour. This reflects the relationship between exposure to the drug and pharmacological response. Assessing pharmacodynamic parameters allows for the determination of whether a drug administered at a particular dose leads to modulation of the target. This is particularly important in phase I studies, where the goal is to assess the feasibility and safety of new therapeutic agents and to define the maximally-tolerated dose. Usually determination of pharmacodynamic effects requires serial tumour sampling through biopsies. However, acquiring tumour tissue can be challenging, depending on the location of the tumour, as well as the invasiveness and associated risks of the biopsy procedure required. A way to indirectly obtain information on target effects is to use surrogate normal tissues. For example, skin, hair follicles, and peripheral blood mononuclear cells (PBMCs) are all sources of normal tissue that are used for pharmacodynamic assessments in early phase clinical trials. However, there are several limitations to this approach, including the lack of expression of oncogenic targets in normal tissues, such as acquired oncogenic somatic mutations, issues with tumour heterogeneity, as well as differences in drug concentrations between surrogate normal tissues and tumour tissues.

Surrogate Endpoints

Surrogate endpoints or biomarkers are often used as an intermediate readout of treatment effect at a point in time earlier than the clinical endpoint of interest. Surrogate markers are typically assessed in situations where there is a long time course to an event that is clinically meaningful, such as survival. For example, pathological complete response (pCR) and progression-free survival (PFS) are often used as surrogate markers for overall survival in breast cancer clinical trials. However, it is important to note that surrogate markers such as pCR and PFS may not always correlate with overall survival, as was shown for bevacizumab in metastatic breast cancer, where a PFS benefit did not translate to improved overall survival.

Another surrogate endpoint is treatment-related toxicity, which is used in monitoring chemotherapy drugs and targeted agents. One example with drugs that affect cell proliferation is myelosuppression. For example, the depth of myelosuppression that occurs with drugs that affect cell proliferation is reflective of the drug's antiproliferative effect, and thus may serve as an early indicator of anti-tumour activity, before radiological response is seen. Another example is hyperglycaemia that is observed with phosphoinositide 3-kinase (PI3K) inhibitors. This is thought to be a mechanism-based toxicity, as the alpha subunit of PI3K has an important role in insulin signalling, and thus it may be potentially used as a surrogate marker of target inhibition.

Conclusions and Future Directions

The sequencing of the human genome and of many solid tumours has identified key genetic alterations and has provided insight into the biological mechanisms involved in oncogenesis. At the same time, advances in molecular biology and histopathology techniques have enabled more detailed characterisation of tumour tissues. Elucidation of important signalling mechanisms deranged in cancer has led to the identification of new drug targets, enabling the development of novel therapies. The use of biomarkers has facilitated the selection of patients who would derive the most benefit from systemic therapy and a more personalised approach to treatment. However, only a few biomarkers for common solid tumours are currently routinely tested in the clinical setting. In the future, the development of less expensive and high-throughput sequencing methods will allow for the expansion of testing for genetic alterations to all tumours and patients and will lead to more tailored therapies. New and less invasive methods for obtaining tumour material such as measurement of circulating tumour cells in peripheral blood will enable more frequent monitoring of tumour response to therapy. Patients will benefit from this individualised approach to cancer care by receiving therapies modified to their unique molecular and cellular characteristics, leading to an improved therapeutic benefit to toxicity ratio. Ongoing biomarker development will allow the oncologist not only to better define prognosis and predict treatment response but also provide an early indicator of treatment efficacy.

A greater understanding of the pharmacodynamic effects of drugs and the use of surrogate biomarkers will also aid clinicians in identifying patients who are not responding to standard therapies and may therefore need a different treatment approach. Additional research is needed to develop new biomarkers to better select patients suitable for specific therapies, to monitor treatment response, and to identify patients at greater risk of toxicities. The incorporation of novel biomarkers in phase I clinical trials may also improve the efficacy of experimental drug testing, through the early identification of patients likely to respond to a new treatment for enrichment in phase II/III trials.

Declaration of Interest:

Dr Pezo has reported no conflicts of interest.

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Further Reading

Ang JE, Kaye S, Banerji U. Tissue-based approaches to study pharmacodynamic endpoints in early phase oncology clinical trials. Curr Drug Targets 2012; 13:1525–1534.

Bedard PL, Hansen AR, Ratain MJ, et al. Tumor heterogeneity in the clinic. Nature 2013; 501:355–364.

Bigbee W, Herberman RB. Tumor Markers and Immunodiagnosis. In: Cancer Medicine, 6th edition. Kufe DW, Pollock RE, Weichselbaum RR, et al, Eds. Hamilton: Decker Inc.; 2003.

Chapman PB, Hauschild A, Robert C, et al. Improved survival with vemurafenib in melanoma with BRAF V600E mutation. N Engl J Med 2011; 364:2507–2516.

Chung C, Christianson M. Predictive and prognostic biomarkers with therapeutic targets in breast, colorectal, and non-small cell lung cancers: a systemic review of current development, evidence, and recommendation. J Oncol Pharm Pract 2014; 20:11–28.

Doroshow JH, Kummar S. Translational research in oncology – 10 years of progress and future prospects. Nat Rev Clin Oncol 2014; 11:649–662.

Hayes DF, Bast RC, Desch CE, et al. Tumor marker utility grading system: a framework to evaluate clinical utility of tumor markers. J Natl Cancer Inst 1996; 88:1456–1466.
Kalia M. Biomarkers for personalized oncology: recent advances and future challenges. Metabolism 2015; 64(3 Suppl 1):S16–S21.
Kelloff GJ, Sigman CC. Cancer biomarkers: selecting the right drug for the right patient. Nat Rev Drug Discov 2012; 11:201–214.
Nalejska E, Maczyńska E, Lewandowska MA. Prognostic and predictive biomarkers: tools in personalized oncology. Mol Diagn Ther 2014; 18:273–284.
Soda M, Choi YL, Enomoto M, et al. Identification of the transforming EML4-ALK fusion gene in non-small-cell lung cancer. Nature 2007; 448:561–566.
Venter JC, Adams MD, Myers EW, et al. The sequence of the human genome. Science 2001; 291:1304–1351.
Vogelstein B, Papadopoulos N, Velculescu VE, et al. Cancer genome landscapes. Science 2013; 339:1546–1558.