The era of a one-size-fits-all approach to the treatment of many diseases was dominant in the 20th century, but is rapidly receding and being replaced with an increasingly personalised ‘precision medicine’ (PM) approach. Biomarkers (also known as a ‘molecular markers’ or ‘signature molecules’) are now being used for pre-disposition, predictive, diagnostic, prognostic, toxicological or monitoring purposes, and patients are benefiting from more individualised treatments. Oncology has led the field in introducing precision therapies, although a similar approach is now being developed in areas such as neurology and endocrinology. In this new genomic era of medicine, healthcare professionals need to have a sound working knowledge of not just the PM available and used in practice, but also of the biomarkers targeted and the pharmacogenomic tests available to detect them.

This review focuses on predictive biomarkers, with some examples of pre-disposition, diagnostic or prognostic biomarkers. It aims to bring together anticancer agents that have been approved for use in the UK (at the time of publication) as part of a PM approach, along with information relating to the relevant biomarkers and biomarker assays available. Although this should prove useful to practicing clinicians and pharmacists, it can only represent a snapshot of the current therapeutic landscape owing to the increasingly rapid movement of the field. The review is in two parts and presented in sections relating to cancer type, although some biomarkers are relevant to more than one.

Part 1 focuses on solid tumours, and includes an introduction to the technologies used for companion diagnostic tests, along with discussions of the limitations of biomarker testing, the role of regulatory bodies in validating biomarkers and companion diagnostic tests, PM-based clinical trials, tumour agnostic anticancer agents and related biomarkers, supportive therapies, and funding challenges for the NHS in relation to the growing introduction of novel PM agents and biomarker assays. Part 2 focuses on haematological cancers, and includes a summary of potential future applications of the PM approach in oncology.

Key words: biomarkers, cancer, clinicians, oncology, personalised medicine, pharmacists, precision medicine.

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Key points

- Introduction of the ‘precision medicine’ approach has seen its most significant advances in the field of oncology.
- The key scientific principle behind this approach is to identify predictive biomarkers that can be used to select the most suitable therapeutic agents.
- Biomarkers can also be used for diagnostic, prognostic, toxicological and treatment monitoring purposes.
- Many pharmacogenomic assays and kits have been developed to identify biomarkers.

Introduction

Each individual has their own unique genome, which can include small single nucleotide polymorphisms (SNPs) and/or large changes in DNA base pair sequence (mutations). These can be inherited but can also be introduced during a person’s lifetime through external agents (e.g. carcinogenic chemicals or radiation). Although usually harmless to the well-being of the individual, these genetic modifications can affect the way the body responds to a therapeutic agent either through differences to the drug target, or through ADMET considerations (i.e. absorption, distribution, metabolism, excretion and toxicology).
Precision medicine, sometimes known as ‘personalised medicine’ and abbreviated to ‘PM’, is a term that is increasingly being used to describe treatments, including therapeutic agents, tailored to individual patients or groups of patients. The overall goal is to match therapies to individuals to ensure that they receive effective treatment with minimal toxicity. This is particularly important for cancer patients who may have a limited life expectancy. Furthermore, there has been an ongoing decrease in the cost of sequencing the human genome, which has led to the widespread adoption of integrative sequencing strategies for the study of cancer and the PM approach.

Although often used interchangeably with personalised medicine, the term ‘precision medicine’ was first introduced by the US National Research Council in 2011 with the aim of conveying a broader concept that, although it may not be possible to develop treatments specifically for individual patients on a one-off basis, it should be feasible to at least define subgroups of patients and target them through a genomics-based approach. The concept of PM in oncology gained further momentum when the PM Initiative was launched in the US in 2015, accelerating the development of biomarker-driven therapeutic strategies.

Within the field of oncology, the most significant aspect of a PM approach involves the identification of a ‘biomarker’ associated with a particular cancer type. A biomarker is a unique mutated nucleic acid sequence, protein, glycoprotein or group of proteins, expressed by the tumour cells but not normally by healthy cells. There are four main types of biomarkers: pre-disposition (indicating the likelihood of developing the disease), diagnostic (used to confirm the patient has a particular cancer), predictive (determining which cohort of patients may benefit from a particular drug therapy) and prognostic (suggesting how the cancer may develop in the individual). Each biomarker type is relevant at different stages of the disease (see Figure 1).

<table>
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<td>Pharmacokinetics (e.g. genetic mutations that prevent metabolism of particular drugs)</td>
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Figure 1: Role of biomarkers in oncology

There have been multiple successes in the PM approach in introducing improved treatments in the oncology area, with imatinib (Glivec; Novartis) generally acknowledged as the first agent of this type. Developed in the 1990s and approved by the US Food and Drug Administration (FDA) in 2001, it targets the ATP-binding pocket of the mutant BCR-ABL protein, which is uniquely found in the tumour cells of chronic myeloid leukaemia (CML) patients but not their healthy cells. Patients are selected for treatment based on the presence of the genetic abnormality BCR-ABL1 Ph+ (i.e. the Philadelphia chromosome) in their tumour cells using an assay, such as Quantidex BCR-ABL (Asuragen Inc) (see Part 2 of this article).
Imatinib and its successors have made a noticeable difference to the lives of CML patients, with simple oral dosing, significantly improved five-year survival compared with traditional cytotoxic therapies and a lower level of side effects leading to a reduction in the need for hospital visits and an improved overall treatment experience. A more recent success is the development of the \( \text{BRAF}^{V600} \) inhibitor vemurafenib (Zelboraf; Roche) for the treatment of melanoma, which was approved by the FDA in 2011. Prior to the discovery of this agent, very few treatments were available for advanced melanoma, mostly involving traditional cytotoxic agents that were largely ineffective and caused significant toxicity. For patients identified as suitable for treatment through an appropriate assay (e.g. the Cobas 4800 \( \text{BRAF}^{V600} \) Mutation Test, Roche), a significant improvement in survival rate with reduced side effects is now possible, along with the other advantages of oral dosing.

However, some researchers and clinicians are skeptical of the PM approach, with the EGFR inhibitor gefitinib (Iressa; AstraZeneca) often quoted as an example of a disappointment from the viewpoint of some clinicians. However, in this particular case, the problem was not the agent itself (which is capable of effective EGFR inhibition) but rather the understanding of the biology of lung cancer, in that EGFR inhibition is (and always was) very successful in those patients with activating mutations, but that was not known at the time. Therefore, this example demonstrates the fact that the use of agents such as gefitinib require a clear understanding of tumour biology. This issue with gefitinib was compounded by the desire of some oncologists to combine tyrosine kinase inhibitors (TKIs) with chemotherapy in a way that was almost bound to fail (and frequently did). This suggests that truly successful PM agents may require both design and patient selection based on predisposing mutated proteins present only in the tumour cells.

Owing to the volume of information, the material is presented in two parts. An introduction to the use and limitations of biomarkers is presented in this manuscript, along with a review of the application of a PM approach to solid tumours. Part 2 will cover application of the PM approach to hematological cancers, along with a summary and review of the potential future applications of the PM approach in oncology.

### Technologies used for diagnostic testing

Companion diagnostic (CDx) tests are based on a number of different platform technologies, each with their own advantages and limitations:

- **In situ hybridisation (ISH):** this uses fluorescent nucleic acid probes to detect, bind to, identify, interpret and map mutations in genes, or regions of genes, that frequently appear only in cancer cells. Most tests of this type have the limitation that only single defined mutations are detected, whereas cancer cells can develop multiple mutations.

- **Immunoassays:** based on antibody/antigen recognition, these assays are capable of detecting a range of biomarkers including those related to toxicity, the levels of active drug metabolites in the blood, and the overall levels of drugs for therapeutic monitoring purposes. In general, serum marker testing kits may be non-specific for different cancer types.
Flow cytometry: this uses a laser- or impedance-based biophysical technology for cell sorting and counting, and biomarker detection is achieved by passing a suspension of cells through an electronic detection device. This allows simultaneous multiparametric analysis of the chemical and physical characteristics of thousands of cells per second. Flow cytometry preserves the cells for further studies but requires multiple fluorophores to differentiate cell types, and this can lead to large amounts of data that can be difficult to analyse and interpret.

Tandem mass spectrometry: this methodology, especially if interfaced with liquid, gas or capillary chromatography systems, can be used to accurately measure the levels of both biomarkers and drugs in tissue samples.

Next-generation sequencing (NGS): this is a high-throughput method for screening short or long DNA sequences, as well as identifying biomarkers that are highly expressed. Although the technology is relatively expensive, its costs are reducing, and it is an increasingly recognised method for diagnosing, predicting risk and classifying different cancer types. For example, the FDA has issued two guidance documents to drive the use of NGS as a diagnostic method for identifying the risk of developing a genetic disease. The first provides CDx test developers with FDA-approved public databases containing clinical evidence that can corroborate the accuracy of NGS-based genomic testing results. The second document offers recommendations to potential manufacturers on how to develop NGS testing, highlighting key elements that the FDA will look for in a pre-market submission when assessing an assay's analytical validity.

These different types of companion diagnostic assays will be further described below in the context of specific cancer types.

Limitations of biomarker testing

The NCI Dictionary of Cancer Terms defines a biomarker as a biological molecule found in blood, other body fluids or tissues that is a sign of a normal or abnormal process, or of a condition or disease; or that may be used to see how well the body responds to a treatment for a disease or condition. In this review, the focus is on biomarkers that can be used to select which patients are most likely to respond to particular anticancer therapies. Although simple in concept, in practice there are a number of difficulties and limitations, some of which have been previously reviewed. The most significant problems and limitations are briefly described below.

Acceptability

Whereas blood samples are relatively easy to obtain from patients with haematological cancers, not all solid tumours are straightforward to biopsy. For example, biopsies can more easily be carried out on tumours near to the skin surface (e.g. skin, bladder, breast, prostate, oesophageal and bowel cancers), but for organs situated deeper in the body (e.g. liver, pancreas, lungs) biopsies normally require major surgery. In terms of risk and tolerability for the patient, blood or urine samples are lowest risk and well tolerated. Solid tumour and cerebrospinal fluid biopsies are higher risk in terms of infections and secondary tissue damage, and are less well-tolerated by patients.

Measurement errors
Within the context of this review, the search for a biomarker is usually made in tumour cells obtained from biopsy material taken from solid tumours, or from blood samples in the case of haematological malignancies. There are a number of potential measurement errors associated with sampling, extraction and analysis. For example, with solid tumours that are comprised of multiple clones of cells at different stages of cancer evolution, it is possible that the small sample of cells removed at biopsy do not contain the relevant biomarkers that may be present in cells at other sites within the tumour mass. This is less of a problem with haematological cancers, where a relatively large blood sample is likely to contain at least some of the biomarker-relevant cancer cells. Other errors can result from imperfect extraction and analytical techniques, especially when working with very small samples from solid tumour biopsies.

Sampling bias

This can occur at different stages of the biomarker measuring process, from selecting patients to storing and measuring samples. For example, there may be a bias in sampling patients whose tumours are more convenient for sampling, and also in sampling parts of solid tumours closer to the surface of the body.

Confounding errors

These can arise through failure to identify either internal (e.g. a patient's weight) or external (e.g. suitable laboratory equipment) factors that can affect the amount of evaluable biomarker reported in a sample.

Role of regulatory bodies validating biomarkers and CDx tests

The development of a CDx assay kit in conjunction with a new drug depends on when the biomarker is first identified\[^{28}\]. If a biomarker is first discovered during the early stages of the patient selection process, then the process of assay development can begin early\[^{28}\]. However, if the biomarker is discovered as a response to therapy, development of the diagnostic assay will generally occur at a later stage\[^{28}\]. In 2016, the FDA released draft guidance on the ‘Principles for codevelopment of an \textit{in vitro} companion diagnostic device with a therapeutic product\[^{29}\]’, which aimed to be a practical guide for companies that are developing treatments that rely on a biomarker test for their use. The FDA has recognised that simultaneous development and launch of a drug and corresponding biomarker assay may not always be feasible, therefore this document provides advice for pharmaceutical companies on this issue\[^{29}\]. Although there are some biomarkers that have been specifically approved by the FDA for use in disease monitoring (e.g. nuclear matrix protein-22 for bladder cancer), these biomarkers are not usually specific enough to be used for general population screening\[^{28}\].

CDx tests are classed as medical devices that provide essential information for the safe and effective use of medicines\[^{29}\]. According to the FDA, their three main roles are: identifying patients who are likely to benefit from a particular therapeutic product, to help identify those who are likely to be at an increased risk of developing side effects from a particular medication, and to monitor and adjust a patient’s response to treatment. In oncology, CDx tests are mainly used for identifying patients who are expressing a particular biomarker to select a suitable treatment, and to identify patients at high risk of developing a side effect from a given drug therapy\[^{26}\].
The FDA approves CDx tests through the Center for Devices and Radiological Health (CDRH), which verifies both the analytical and clinical validation of in vivo testing kits. Within the EU, regulation of medicinal devices has been separated from the regulation of pharmaceuticals. The EMA has taken a less active role in regulating companion diagnostics, as the kits are regarded as in vitro diagnostics (IVDs). Therefore, the EMA does not have complete authority over their regulation. According to European directive 98/79/EC, CDx tests that are considered to be an IVD medicinal device and have the ‘CE’ mark can be distributed to all members of the European Economic Area. However, the EU is planning a substantial reform of their current regulation rules to strengthen the system to incorporate new CDx tests into the regulatory framework. Recently, the EMA launched a consultation paper on the development and lifecycle of PMs and their CDx tests.

**Precision medicine-based clinical trials**

To evaluate the PM approach more thoroughly, and to attempt to match targeted therapies to genomic profiling, several clinical trials have been launched in the US. These have been reviewed by Gong et al. and include the Signature, I-SPY, National Cancer Institute (NCI)-sponsored NCI-MATCH, NCI-MPACT, ALCHEMIST, Lung-MAP, Pediatric MATCH, Exceptional Responders, Lung Cancer Mutation Consortium (LCMC)-sponsored, and the American Society of Clinical Oncology (ASCO)-sponsored TAPUR (Targeted Agent and Profiling Utilization Registry) clinical trials. These non-randomised clinical trials are considered to be important for the future of PM approaches, and are aimed at evaluating the performance (both efficacy and safety) of FDA-approved targeted anticancer agents prescribed for the treatment of patients with advanced cancer and with a potentially actionable genomic alteration.

According to ASCO, more than 1,700 participants have consented to participate, and more than 1,220 have been treated with a TAPUR study drug. Clinicians who order the 596-gene Tempus xT assay (Tempus Labs, Inc) receive a report that has been optimised for TAPUR participation with a specific summary of the genomic alterations targeted by study drugs, which helps clinicians screen for trial-eligible patients. Importantly, a number of pharmaceutical companies collaborated to contribute to these trials, and the choices of genomic profiling tests requested by participating clinical oncologists are catalogued with the aim of identifying signals of drug activity. For example, a molecular aberration such as $\text{BRAF}^{V600E}$ seen in any cancer type can be matched for treatment with $\text{BRAF}$ inhibitors (which are presently only FDA-approved for melanoma and a few other specific histologies).

In another example, clinical trials are identifying tumours of any type harbouring mutations in any of the Fanconi anaemia pathway genes, most commonly found in tumours with a mutation pathway in $\text{BRCA1/2}$, to match patients with PARP inhibitors. In the UK, Innovate UK recently launched a €6m initiative for the development of PM technologies that can be used to lead and improve targeted therapies.

**Sources and selection criteria**

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Cancer-relevant biomarkers, the assays to detect and evaluate them, and the associated anticancer agents are reviewed below based on information from the primary and patent literature, and from material available from conferences and the websites of pharmaceutical companies, medical charities and organisations such as the Medicines and Healthcare products Regulatory Agency (MHRA), the European Medicines Agency (EMA), the FDA, the UK government’s National Institute for Health and Care Excellence (NICE)\textsuperscript{39,40} the British National Formulary (BNF), the Journal of Precision Medicine, and web-based resources such as GenomeWeb. Although the information has been grouped in sections relating to cancer type, some biomarkers are highly specific for a particular type (e.g. BCR-ABL for chronic myeloid leukemia), whereas others are associated with more than one tumour type (e.g. BRCA1/2 in breast, ovarian and prostate cancers)\textsuperscript{41}.

Biomarkers, CDx tests and PM anticancer therapies in current use

This part of the two-part review will focus on solid tumours, which are defined as abnormal masses of tissue that do not contain any cysts or liquid areas\textsuperscript{42}, and are classified histologically into sarcomas, carcinomas or lymphomas\textsuperscript{43}. They can be further classified molecularly through the use of processes such as genome sequencing, gene-expression profiling and miRNA analysis, along with other parameters to assess them, for example DNA copy number, chromosomal instability and gene functionality\textsuperscript{44}.

Breast cancer

This is a heterogeneous disease with varied morphological and molecular characteristics\textsuperscript{45}. One of the best ways of distinguishing between the different types of breast cancer is still through histological assessment\textsuperscript{45}. Breast cancers are initially characterised as invasive or non-invasive and, can be further subdivided into carcinomas of no special type (ductal carcinoma) or of a special type, which includes lobular growth patterns and altered differentiation\textsuperscript{45}. Lobular breast cancer in situ, in particular, can be detected through mammography\textsuperscript{45}. Women are offered surgery, radiotherapy or drug treatments (or a combination of one or more of these) as part of their care plan. Biomarkers are increasingly important in the diagnosis, prognosis and treatment of breast cancer, and the most significant ones are summarised in Table 1 along with the CDx tests available and the associated anticancer agents that can be selected on the basis of the presence of a particular biomarker. Progress in this area continues, and recent research has suggested that it may be possible to identify SNPs specific for either lobular or oestrogen receptor (ER)-positive ductal breast cancers\textsuperscript{46}.

The first biomarker to be linked to breast cancer was the estrogen receptor (ER), which was first identified in the 1960s and has been used in the clinical management of this disease since the mid-1970s for diagnostic, prognostic and predictive treatment purposes. ER-positive tumours comprise up to 75% of all breast cancers diagnosed, representing 65% and 80% of patients aged under and over 50 years respectively\textsuperscript{47}. It is also most commonly found in patients with invasive ductal cancers. Both ER and progesterone receptor (PgR) status are routinely assessed for all invasive breast cancers using IHC methods, and the results reported in a standardised way known as the Allred system. This method scores cells based on an estimated proportion and intensity of staining on a scale of 0 to 8, where a score of 3 or greater is defined as positive\textsuperscript{48}. Patients with ER/PgR-positive breast cancer are treated with endocrine therapies such as selective estrogen-receptor modulators (SERMs) (e.g. tamoxifen) or aromatase inhibitors (e.g. anastrazole) in both the adjuvant and metastatic settings.
PgR is used as a predictive biomarker for ER-α function and breast cancer prognosis. In the presence of an agonist ligand, PgR becomes associated with ER-α, directing chromatin-binding events within breast tumour cells, and both receptor and hormone levels can influence the interaction between ER-α and PgR. This cross-talk is involved in regulating the gene expression programme associated with low tumourigenicity, and so is associated with a more favourable outcome for the patient. Although this is a potentially useful biomarker in breast cancer, NICE guidelines state that it should not be used routinely to assess the prognosis of patients with invasive breast cancers. IHC assays are used to assess the level of PgR in tumours.

Gene amplification, or protein over-expression of human epidermal growth factor receptor 2 (HER2), discovered in the 1980s, is often associated with poor prognosis in breast cancer. However, it can be used to identify patients who may benefit from targeted therapies such as the antibody trastuzumab (Herceptin, Roche) or the antibody–drug conjugate (ADC) trastuzumab emtansine (Kadcyla, Roche). HER2 is over-expressed in around 15% of cases of early invasive breast cancer, and around 77% of those that are also ER-positive. Diagnostic tests for HER2 over-expression and gene amplification include IHC and ISH methods, and standardised quality-assured methodologies have been developed. The results are reported as HER2 negative or HER2 positive according to NICE guidelines. The results are reported as HER2 negative or HER2 positive according to NICE guidelines, with test results scoring 3+ by IHC, or 2+ and amplified by ISH, counted as positive.

Mutated versions of the breast cancer genes 1 and 2 (BRCA1 and BRCA2), although found in only a small percentage of the general population, are classified as high-risk mutated genes, and are the best-known predictive genetic biomarkers for both breast and ovarian cancer. BRCA1 and BRCA2 are tumour suppressor genes that have been shown to be pivotal in many cellular processes, including the transcriptional regulation of DNA after damage, as well as DNA repair. In addition, they are involved in protecting the genome from damage through the BRCA proteins that maintain chromosomal stability. Mutations in these genes can be passed through generations in both sexes. However, a recent study has suggested that young breast cancer patients with BRCA mutations have a similar survival rate to those without the mutation. This is an important finding as, prior to this study, early diagnosis of this mutation frequently led to elective double mastectomies in healthy patients as a prophylactic measure, and in risk-reducing bilateral salphingo-oophrectomy in ovarian cancer patients after chemotherapy treatment.

The CDK4 and CDK6 kinases associated with Cyclin D1 are over-expressed in all subtypes of breast cancers. The presence of both CDK4 and Cyclin D1 are needed to maintain tumour cell proliferation. While the Cyclin D1/CDK4 complex has two functions, the main one is to sustain the tumourigenic potential of breast cancer cells, although researchers agree that more studies are required to fully explain why the Cyclin D1/CDK4 complex plays a critical role in tumour cells but is less important in normal cells. Drugs such as palbociclib (Ibrance, Pfizer), ribociclib (Kisqali; Novartis) and abemaciclib (Verzenio; Eli Lilly) are small-molecule inhibitors of CDK4 and CDK6, with a high selectivity for these kinases compared with other cyclin-dependent kinases. Although no CDx tests are available, for these agents, they appear to work more effectively in patients with hormone-receptor-positive breast cancer, and are synergistic with other endocrine therapies.
PIK3CA mutations with associated over-activation of the PI3K signalling pathway were identified by Novartis scientists as associated with tumour growth, resistance to endocrine-based therapies and relatively poor treatment outcomes\cite{63,64}. A recent study (i.e. SOLAR 1) analysed breast cancer tissue and circulating tumour DNA in 572 patients, identifying PIK3CA mutations in 40% of hormone receptor positive or negative breast cancers\cite{63,64}. The kinase inhibitor alpelisib (Piqray) was developed by Novartis to target PIK3CA mutations, and is approved by the FDA for use in breast cancer\cite{63,64}. It is used alongside Qiagen’s Therascreen PIK3CA RGQ PCR test to select patients suitable for treatment\cite{63,64}. Development of a similar kinase inhibitor (i.e. taselisib) by Roche to target PIK3 mutations for the treatment of metastatic breast cancer was halted due to a lack of efficacy and unacceptable toxicity\cite{65}.

In addition to OncotypeDX, other test kits that profile panels of genes to assist with clinical decision making in breast cancer include MammaPrint (Agendia), EndoPredict (Myriad Genetics Inc), IHC4, Mammostrat (Clariant Diagnostic Services) and Prosigna (NanoString Technologies Inc). However, none of these have been recommended for use in the UK by NICE\cite{66}, which has produced several draft reports weighing evidence on the use of diagnostic testing kits of this type\cite{67}.

See Supplementary PDF for Table 1: Biomarkers important in breast cancer, the PM agents used in this disease, and the companion diagnostic tests used to select patients most likely to respond

Sources: \cite{66,68,69,70,71,72,73}

Lung cancer (small cell and non-small cell)

EGFR plays a critical role in regulating normal cell proliferation, apoptosis and other cellular functions. Around 10% of non-small cell lung cancer (NSCLC) patients in the UK and 35% in East Asia have EGFR mutations associated with their tumours\cite{74}. Initial clinical studies carried out by Riley et al. showed that first-generation tyrosine kinase inhibitors (TKIs) such as gefitinib and erlotinib gave a significant clinical response in 10% of Caucasian and 25–30% of Asian patients\cite{75}. However, patients who responded well initially soon developed resistance due to secondary acquired EGFR mutations (75), the main one of which was a change of threonine to methionine at amino acid position 790 (i.e. T790M) in exon 20 of the EGFR gene\cite{76}, resulting in a reduced binding affinity for these first-generation TKIs\cite{76}. This understanding of the mechanism of action allowed researchers to develop improved TKI molecules, such as osimertinib (a third-generation TKI)\cite{76,77}. A number of companion diagnostic tests have been developed to identify patients with EGFR biomarkers, and some of these have been approved for use in the NHS (see Table 2).

**EML4-ALK** (echinoderm microtubule-associated protein-like 4 — anaplastic lymphoma kinase) is a mutated translocation fusion gene resulting from an inversion within the short arm of chromosome 2 where ALK joins with EML4\cite{78}. Patients who are ALK+ can be treated with TKI agents specifically designed to target this mutation. The first-generation of these was crizotinib (Xalkori; Pfizer), although further generations of ALK-1 inhibitors have since been developed.
ROS-1 is a receptor tyrosine kinase encoded by the ROS-1 gene. It has structural similarity to the ALK protein but the roles of the ROS-1 protein in normal development, and the identity of its physiologic ligand, are presently uncertain. It is classed as an oncogene because it can rearrange into the ROS-1 fusion mutation, which is commonly seen in young non-smoking female patients of Asian ethnicity with adenocarcinoma histology. Crizotinib has been approved by the FDA and NICE as a viable therapy for patients whose tumours have a ROS-1 translocation\cite{78}. CDx tests for both ALK and ROS-1 biomarkers are approved for use in the NHS (see Table 2).

Vascular endothelial growth factor (VEGF 1–3), platelet-derived growth factor (PDGF) and fibroblast growth factor (FGF) are all secreted by both normal and tumour cells, and induce angiogenesis by specifically binding to their respective receptors\cite{79}. Angiogenic pathways are very important for the development of NSCLC as they promote tumour progress by facilitating the growth of new blood vessels\cite{74}. Therefore, these three growth factors, which are involved in the regulation of angiogenesis, can be used as biomarkers for drug discovery and patient selection. The VEGF inhibitor bevacizumab (Avastin; Roche) was one of the first agents to be developed in this area, and is licenced for use in NSCLC in combination with a platinum-based therapy for the first-line treatment of NSCLC tumours harbouring VEGF as a biomarker. However, it is not recommended by NICE for this purpose based on a lack of evidence (NICE Technical Appraisal TA148)\cite{74}.

PDGF plays an important role in the development of normal cells, and is essential for cell proliferation, migration, differentiation and survival. There are two main receptor isoforms (i.e. α and β), and both are associated with poor prognosis in lung cancer. It is also involved in angiogenesis, and regulates VEGF expression in tumour cells, which can progress the development of pre-cancerous NSCLC into a more advanced stage. One of the most effective ways of blocking PDGF activity is through the inhibition of intracellular PDGFR kinases.

The FGF family of growth factors is involved in numerous cellular processes, one of which is angiogenesis\cite{80}. FGF-2 is one of the most potent angiogenic factors within the family, and high expression of this ligand is associated with transformation of normal cells into malignant ones\cite{80}. The specific role of FGF-2 in NSCLC development and progression is largely unknown\cite{80}, although one study has established a potential role of FGFR-dependent signaling as a component of the resistance mechanism for EGFR tyrosine kinase inhibitors\cite{81}. Both FGFR and VGF are structurally similar, and so novel inhibitors of both are at the research stage as targeted anti-angiogenesis agents\cite{81}. FGFR inhibitors could potentially be used either alone or in combination with other TKIs as a means of expanding the portfolio of therapies that can be used for NSCLC patients\cite{81}.

The BRAF\textsuperscript{V600E} and BRAF\textsuperscript{V600K} biomarkers (important in melanoma — see ‘Metastatic malignant melanoma’) are present in 1–2% of lung adenocarcinomas in patients who do not have common lung tumour mutations such as EGFR, EML4-ALK and ROS-1\cite{82}. The Oncomine Dx Target Test (Thermo Fisher Scientific) based on next-generation sequencing was approved to test for BRAF, ROS-1 and EGFR mutations in these patients\cite{82}. NGS panels have also been used in patients with NSCLC to detect a range of genomic alterations, including the BRAF\textsuperscript{V600E/K} mutations\cite{84}. 
There has been some success in measuring PD-L1 over-expression in lung tumours to predict the possible outcome of treatment with the immunotherapy agent pembrolizumab (Keytruda; MSD). However, the same predictive results have not been observed with nivolumab (Opdivo; Bristol-Myers Squibb). Despite the relative lack of success with this biomarker, for some patients the results are dramatic and can lead to immunotherapy supplanting all other treatments. At the time of writing, the FDA has expanded use of the approved Dako PD-L1 IHC 22C3 PharmDx assay (the CDx assay for pembrolizumab) from NSCLC to a variety of cancer types including gastric, gastroesophageal junction adenocarcinoma and cervical cancer. However, in other tumour types (e.g. urological malignancies), PD-L1 over-expression does not appear to have any predictive ability for the benefit of immunotherapy. Findings from the pivotal Phase III KEYNOTE-189 study showed that using pembrolizumab in combination with the antimitabolite pemetrexed (Alimta; Eli Lilly) and a platinum-based agent such as cisplatin or carboplatin significantly improved overall survival, decreasing the risk of death by >50% compared with the use of chemotherapy alone. In the UK, hospitals are currently providing initial triple therapy with platinum-based agents, pemetrexed and pembrolizumab to treat non-squamous NSCLC, followed by binary treatment with pembrolizumab and pemetrexed (as approved by the Cancer Drug Fund).

Tumour mutational burden (TMB) and microsatellite instability-high (MSI-H) are new biomarkers that are being studied for use in personalised immunotherapy treatments. While they are not yet approved as biomarkers for treatment selection, researchers are investigating the use of both to help clinicians accelerate their decisions in ruling in or out treatment options. For example, Bristol-Myers Squibb has a collaboration with Foundation Medicine Inc to use the FoundationOne CDx assay (Roche Products Ltd) to evaluate the use of TMB as a biomarker to assess the efficacy of a combination of two immune-oncology agents, nivolumab and ipilimumab (Yervoy, Bristol-Myers Squibb).

The FoundationOne CDx assay, which was approved by the FDA in 2017, profiles MSI-H and TMB as well as 324 genes, and is approved to help identify best responders to 15 FDA-approved treatments including the anti-PD-1 immunotherapy agent pembrolizumab across five tumour types. Results from the CheckMate-227 trial suggested that advanced NSCLC patients who are expressing high levels of TMB benefit from the nivolumab/ipilimumab combination, with higher progression-free survival observed in patients randomised to the combination arm compared with those given a single agent regardless of their PD-L1 status, suggesting that it may become a first-line therapy option in the future. Bristol-Myers Squibb is currently using the FoundationOne CDx assay to evaluate the use of TMB as a predictive biomarker for nivolumab.

It should be noted that other more-general biomarkers such as vascular endothelial growth factor receptor (VEGFR) and platelet-derived growth factor receptor (PDGFR) have been identified as relevant to lung tumours in some studies.

See Supplementary PDF for Table 2: Biomarkers important in lung cancer, the PM agents used in this disease, and the companion diagnostic tests used to select patients most likely to respond

Source: [87],[91],[92],[93],[94],[95],[96],[97],[98],[99],[100]

Metastatic colorectal cancer and other gastrointestinal tumours
The epithelial growth factor receptor (EGFR), a transmembrane ligand-induced receptor, is a predictive and prognostic biomarker over-expressed by tumour cells in around 70% of patients with mCRC. Anti-EGFR monoclonal antibodies (mAbs), such as cetuximab and panitumumab, work by competitively inhibiting EGFR, preventing its interaction with endogenous ligands with subsequent moderation of downstream signaling pathways including RAS-RAF-MAPK and PI3K-AKT-mTOR.

In some cases, mutations in KRAS, NRAS and BRAF signaling proteins downstream of EGFR may render colorectal cancers unresponsive to anti-EGFR treatment. KRAS genes are among the most frequently activated oncogenes in mCRC. For example, mutations of Kristen RAS (KRAS) occur on exon 2, 3 and 4 in a ratio of 9:1, and are considered to be useful predictive biomarkers for mCRC. KRAS, NRAS and BRAF all predict resistance to anti-EGFR therapies and therefore highlighting their clinical benefit and importance as biomarkers. For example, in a study by Therkilsden et al., only 40–60% of patients with KRAS mutations responded to treatment. Therefore, RAS mutation testing is now carried out to select the most likely responders to anti-EGFR treatment by identifying patients with mutations in signaling proteins including KRAS, BRAF and NRAS. Laurent-Puig et al. have suggested that BRAF mutation status not only predicts the benefits of using anti-EGFR mAbs, but also acts as an adverse prognostic biomarker for the course of mCRC. KRAS mutations can be detected using a range of different techniques (see Table 3) including PCR (e.g. Cobas [Roche], Therascreen [Qiagen] and Idylla [Biocartis]), pyrosequencing, NGS (which allows the measurement of many mutations at once) or IHC, although the latter suffers from limited sensitivity and specificity.

Human Epidermal Growth Factor Receptor 3 (HER3) over-expression has been established as a biomarker in patients harbouring the wild-type KRAS gene who are likely to benefit from anti-EGFR therapy such as panitumumab. Results from the randomised PICCOLO trial revealed that patients with wild-type RAS together with increasing levels of HER3 expression, in addition to high levels of EGFR ligands such as epieregulin and amphiregulin, were associated with prolonged progression-free overall survival when given a combination of panitumumab and irinotecan compared with irinotecan alone. Although not presently tested for, in the future it may be possible to identify mCRC patients who may benefit from an anti-EGFR therapy based on the status of the HER3 and EGFR ligands together.

The angiogenic biomarker vascular epithelial growth factor (VEGF) is associated with progression, invasion and metastasis of CRC. It has been demonstrated that over-expression of VEGF mRNA in a primary tumour is highly correlated with a poor prognosis. Therefore, this biomarker is potentially useful prognostically when diagnosing the stage of CRC. In the same study it was found that hypoxia can activate the expression of VEGF in CRC tumour cells, thus promoting angiogenesis.

UDP-Glucuronosyltransferase 1A1 (UGT1A1) is an enzyme involved in the glucuronidation of SN-38, an active metabolite of irinotecan (Campto; Pfizer). It is a proven safety biomarker for this treatment in mCRC patients harbouring a polymorphism of the UGT1A1 gene (i.e. UGT1A1*28). This polymorphism is associated with a high risk of grade 3 or 4 haematological toxicity through decreased metabolism of SN-38. Thus, checking the UGT1A1 status of patients before initiating irinotecan therapy, and starting with a lower dose if the mutation is present, is recommended but not obligatory in many health systems globally. Testing is not put into practice by most oncologists, and the test is not routinely available in the NHS.
PD-L1 expression associated with high microsatellite instability (MSI-H; Microsatellite Instability-High) or dMMR (Mismatch Repair Deficiency) biomarkers in CRC tumour cells can be used to identify patients who may benefit from immuno-oncology agents such as pembrolizumab or nivolumab[113]. Although pembrolizumab does not currently have an established role in the treatment of mCRC, in mid-2017 the FDA granted accelerated approval for the use of pembrolizumab in patients with unresectable or metastatic solid tumours with MSI-H or dMMR biomarkers. This was the first time that the FDA had accelerated a drug approval based on the genomic features of a tumour rather than its originating tissue or organ[114][115] (see later). There are two clinically useful tests to detect MSI-H/dMMR biomarkers in cancer patients. The first involves the identification of MSI-H directly by molecular evaluation of poly-A microsatellites. The second is based on demonstration of lack of expression of MMR proteins through immunohistochemistry, which provides an indirect indication of an abnormal dMMR system[46].

The biomarkers, companion tests and associated drug treatments for mCRC are summarised in Table 3 below.

See Supplementary PDF for Table 3: Biomarkers important in metastatic colorectal cancer (CRC), the PM agents used in this disease and the companion diagnostic tests used to select patients most likely to respond

Source:[87][98][116][117]

A recent study at the Mayo Clinic has uncovered several DNA mutations in colorectal cancer-adjacent polyps (CAPs) that differentiate them from cancer-free polyps (CFPs)[118][119]. It is anticipated that this discovery may help clinicians tailor the colonoscopy surveillance interval for individual patients depending on the type of polyps found[118][119].

Other tumours of the gastrointestinal tract may also be amenable to a PM approach in the future[108]. For example, Ploidy testing is carried out to support a diagnosis of Barret’s oesophagus, and is used to guide endoscopic surveillance although it does not currently have treatment implications. Furthermore, Lucid Diagnostics Inc is commercialising EsoCheck, a squamous cell-based test used to detect Barret’s oesophagus[120]. EsoCheck utilises a balloon sampling device that allows cells from the surface of the oesophagus to be collected and checked for DNA methylation of the VIM and CCNA1 genes as biomarkers for the disease. Early trials have suggested that the test may be >90% accurate in identifying patients with Barret’s oesophagus.

Metastatic malignant melanoma
Around 50% of melanoma patients have *BRAF* mutations in their tumour cells\[121\] (see Figure 2)\[122\]. More than 90% are within codon 600 and, of these, more than 90% are a single nucleotide mutation in the kinase protein resulting in the change of a valine (V) at amino acid position 600 to a glutamic acid (E)\[121\]. This mutation, known as *BRAF*\(^{V600E}\), causes melanoma cells to proliferate aggressively. The first agent designed to target this mutated kinase, vemurafenib (Zelboraf; Roche), discovered by Plexxikon Inc but now owned by Daiichi-Sankyo/Genentech, proved very successful in the clinic and was approved by the FDA in 2011 for the treatment of late-stage melanoma. Another anti-*BRAF*\(^{V600E}\) agent, dabrafenib (Tafinlar), followed for the treatment of advanced melanoma, and was initially approved by the FDA in 2013 as a single agent treatment for patients with *BRAF*\(^{V600E}\) mutation-positive disease. Vemurafenib is significant in being the first drug to be discovered through a fragment-based lead discovery approach to gain regulatory approval. A PCR test for the *BRAF*\(^{V600E}\) mutation is used to determine whether patients are suitable for these agents (see Supplementary PDF for table 4).

Interestingly, according to a recent study carried out by da Silva et al.\[123\], melanoma patients with *BRAF*\(^{V600K}\) mutations appear to benefit less from *BRAF* and *MEK* inhibitors than those whose tumours contain *BRAF*\(^{V600E}\) mutations\[123\]. The authors suggest this may be due to less reliance by the tumour cells on ERK pathway activation and the greater use of alternative pathways\[123\]. However, in contrast, the patients with *BRAF*\(^{V600K}\) mutations have a higher mutational load in their melanoma cells and seem to respond better to immunotherapy\[123\].
MEK (MAPK/ERK kinase) is the first kinase downstream from BRAF in the EGFR signaling pathway (see Figure 2). The development of MEK inhibitors was based on the rationale that they should block signaling in this pathway when used alone, but should also be synergistic with BRAFV600 inhibitors as the two adjacent kinases could be blocked simultaneously, thus further reducing the possibility of signaling, and with the pathway still inhibited even if resistance develops toward one of the agents. Trametinib (Mekinist, Norvartis), was the first agent of this class to be approved by the FDA. In pre-clinical in vitro and in vivo studies, MEK inhibitors were shown to block the growth of melanoma cells carrying the BRAF\textsuperscript{V600E} mutation\cite{122} and, in patients, two key clinical trials (i.e. the COMBI-D and COMBI-V trials) evaluated the combination of anti-MEK and anti-BRAF agents. For a combination of dabrafenib and trametinib, a 29% and 31% reduction of risk of death was obtained compared with the use of dabrafenib or vemurafenib alone, respectively, and the quality of life measure also improved\cite{124,125}. A second MEK inhibitor, cobimetinib (Cotellic, Roche), followed from Exelixis/Genentech and was approved by the FDA in 2015 for use in combination with vemurafenib for unresectable or metastatic melanoma with BRAF\textsuperscript{V600E} or BRAF\textsuperscript{V600K} mutations.

More recently it has been discovered that the CTLA-4 and PD-1 immune checkpoint pathways, which down regulate T-cell activation to maintain peripheral tolerance, play an important role in melanoma as well as other tumour types. Melanoma cells have been shown to exploit both checkpoint pathways to induce an immunosuppressive state that allows the tumour to survive and grow instead of being eliminated by the immune system\cite{126}. Therefore, anti-CTLA-4 agents (e.g. ipilimumab) or anti-PD-L1 agents (e.g. pembrolizumab, nivolumab or atezolizumab), otherwise known as immune-oncology agents or checkpoint inhibitors, are now used in the treatment of metastatic melanoma (see Table 4). Interestingly, a recent study described a common mechanism of intrinsic and acquired resistance to these therapies associated with mutations in a gene coding for β-2-microglobulin, a contributor to the Class I antigen presentation by the major histocompatibility complex\cite{115}. Discovered through exome sequencing of tumour samples taken from 17 affected patients, this could point the way to a pharmacogenomic-based test to determine which melanoma patients are most likely to benefit from immune-oncology treatment.

See Supplementary PDF for Table 4: Biomarkers important in metastatic melanoma, the PM agents used in this disease and the companion diagnostic tests used to select patients most likely to respond

Source:\cite{91},\cite{92},\cite{93},\cite{127}

In 2013, the FDA approved the use of dabrafenib and trametinib in combination alongside the THxID BRAF test from BioMérieux. It is the second companion diagnostic approved by the FDA for BRAF mutation detection following approval of Roche's Cobas 4800 BRAF V600 Mutation Test in 2011.

Understanding the different genotypic mutations that occur in patients with melanoma is now considered a crucial part of treating this disease\cite{128}, particularly as identification of a specific mutation (e.g. BRAF\textsuperscript{V600E}) can lead directly to highly effective targeted therapy. Tests such as the Cobas 4800 BRAF V600 Mutation Test and THxID-BRAF CDx Test (see Table 4) have been developed to identify this one single mutation. However, other tests such as the Oncomine Dx Target Test and NGS tests such as the Ampliseq Hotspot Panel for melanoma and the IonTorrent Personal Genome Machine\cite{129,130} have the advantage of providing mutation information across a number of genes simultaneously, thus potentially providing options for other treatments\cite{128}. 

17/66
Ovarian cancer

Around 10% of invasive ovarian cancers have a hereditary basis, and many of these involve mutations in the **BRCA1** and **BRCA2** genes. Based on data from the Royal Marsden Foundation\[^{131}\]; patients who carry a mutation in **BRCA1** have a 40–60% chance of developing ovarian cancer, with the risk increasing after the age of 40. Those with **BRCA2** mutations have a lower risk (i.e. 10–30%), although this also increases with age from the mid-late 40s.

Patients with ovarian tumours containing **BRCA1** and **BRCA2** mutations tend to demonstrate higher platinum sensitivity over a longer period of time compared with those with no **BRCA** mutations. They also have an improved response to the PARP (Poly ADP Ribose Polymerase) inhibitors niraparib (Zejula; Tesaro UK Ltd), olaparib (Lynparza; AstraZeneca) and rucaparib (Rubraca; Clovis Oncology UK Ltd)\[^{131};\[^{132}\]. PARP inhibitors were developed through research on the different mechanisms of DNA single-strand break repair, through either homologous recombination or non-homologous end joining\[^{133}\]. Inhibition of either one of these mechanisms separately has no effect on a tumour cell, whereas inhibition of both pathways simultaneously becomes a lethal event. Alongside Myriad Genetics’ BRCAnalysis CDx for olaparib and niraparib, a diagnostic test from Foundation Medicine has recently been approved by the FDA to identify patients suitable for treatment with rucaparib\[^{134};\[^{135}\]. More recently, AstraZeneca has been working with Myriad Genetics, using its MyChoice HRD Plus genetic test to select patients for treatment with a combination of olaparib and the anti-angiogenic agent bevacizumab (Avastin; Roche) (see Table 5)\[^{136}\]. This test works by identifying and detecting patients with somatic mutations in their **BRCA1/2** genes, as well as profiling 102 other genes\[^{136}\]. It also identifies other mutagenic changes, such as loss of heterogeneity, telomeric allelic imbalance and large-scale state transitions\[^{136}\].

Angiogenesis is a highly dynamic process regulated by pro- and anti-angiogenic modulating ligands, of which VEGF is one\[^{137}\]. VEGF is considered to be the predominant growth factor expressed by tumour cells, and is regulated by internal factors such as hypoxia, acidosis, mechanical stress, and alterations in oncogenes and tumour suppression genes\[^{137}\]. Over-expression of VEGF occurs in many solid tumour types, and is associated with shortened survival in ovarian cancer\[^{137}\]. VEGF is expressed in more than 70% of ovarian cancers, and its level of expression is correlated with the stage of malignancy\[^{137}\]. VEGFR-2 has been found in the tumour cells of 75% of ovarian cancer patients, and its inhibition is associated with a reduction in tumour growth and vascularisation in in vivo experiments, highlighting its importance in ovarian cancer prognosis and treatment\[^{137}\]. The anti-angiogenic agent bevacizumab (Avastin), in combination with paclitaxel and carboplatin, has been approved by NICE for use in advanced disease as a first-line treatment\[^{137};\[^{126}\].

Other treatments for ovarian cancer include farletuzumab (MORAb-003), a monoclonal antibody that targets FR-alpha (a biomarker over-expressed in some cancers including ovarian cancer), which is presently being evaluated in combination with carboplatin and taxane in Phase III clinical trials in patients with relapsed platinum-sensitive ovarian cancer\[^{138}\]. If successful, FR-alpha could be used in the future as a biomarker to select patients who may benefit from treatment with this agent.

Clinically, ovarian cancer is usually diagnosed through an ultrasound in conjunction with measurement of the blood level of the CA125 protein, although diagnosis through tissue biopsy remains the gold standard, owing to the wide spectrum of characteristics of epithelial ovarian tumours. Morphotek Inc and Fujirebio Diagnostics are collaborating to develop, validate, manufacture and commercialise a diagnostic kit to detect CA125 II, a biomarker that is highly expressed in benign, borderline and malignant epithelial ovarian tumours\[^{139}\].
On the basis of Phase III results, an anti-angiogenic gene therapy agent, ofranergene obadenovec (developed by Vascular Biogenics Ltd), was awarded Orphan Drug Case designation by the EMA in 2017. This agent encodes a fusion protein that combines the extracellular and intra-membranal domains of human tumour necrosis factor 1 with a Fas receptor that contains a hypoxia-responsive element capable of driving cell death in the endothelium of blood vessels following expression of the chimeric protein.

See Supplementary PDF for Table 5: Biomarkers important in ovarian cancer, the PM agents used in this disease and the companion diagnostic tests used to select patients most likely to respond.

Source: [68], [69], [70]

Prostate cancer

Prostate cancer occurs most commonly in men aged >65 years, particularly in those of African descent. It has also been established that African Americans with prostate cancer who express protein isoforms such as PIK3CD, FGFR3, TSC2 and RASGRP2 do not always respond to targeted therapies, potentially explaining the higher mortality rate in this population.

The best-known biomarker for prostate cancer is PSA (prostate-specific antigen), a protein produced by the prostate gland and typically over-expressed when the prostate becomes enlarged, inflamed or contains tumour cells. PSA can be detected using a simple blood test, but has not yet been successfully targeted for the development of novel therapeutic agents. Instead, clinicians use PSA levels as part of a male health screen to trigger further investigations. Even though the FDA has approved the use of PSA testing as a screening tool for diagnosing prostate cancer, the test is non-specific because PSA levels can increase in both benign prostatic hyperplasia (BPH) and in prostate cancer, which leads to a high false-positive rate for prostate cancer. Conversely, PSA expression can be low or normal in some cases of small cell prostatic carcinoma. At present, magnetic resonance imaging (MRI) is the gold standard for detecting prostatic carcinoma, which is then followed by a targeted biopsy if necessary.

There are a number of treatment strategies and agents used to treat prostate cancer, but most are not directly linked to PSA levels. However, in early 2018 the FDA approved a novel agent apalutamide (Erleada ) for patients with non-metastatic prostate cancer resistant to standard hormone therapy (i.e. androgen deprivation therapy). In SPARTAN, a Phase III clinical trial for this agent that led to FDA approval, patients with castration-resistant prostate cancer but no metastatic disease were selected based on rapidly rising PSA levels (e.g. short PSA doubling time). They were randomly assigned to receive apalutamide or placebo in addition to ongoing androgen deprivation therapy, with a 70% reduction in mortality rate observed in the apalutamide treatment group. At present, patients are not specifically selected for treatment with apalutamide based on PSA levels, although research on this correlation is continuing.
Mutated BRCA1 and BRCA2 genes are also becoming important biomarkers for prostate cancer, and have been used to select patients for therapy with PARP inhibitors such as olaparib\textsuperscript{134}. In the TOPARP-A trial, blood samples from a small cohort of men with advanced prostate cancer were checked for DNA containing BRCA1 and BRCA2 mutations in the blood\textsuperscript{131,144}. The trial consisted of six patients who were selected to have homologous recombination deficiency (HRD) somatic mutation. The results suggested that HRD testing could be used to identify patients suitable for treatment with PARP inhibitors such as olaparib\textsuperscript{134}. Other novel biomarkers linked to HRD have been found in 30\% of patients with lethal prostate tumours\textsuperscript{134}, although these have not yet been used for drug discovery purposes.

Interestingly, studies have suggested that in prostate cancer patients carrying the BRCA2 mutation, the order in which some anticancer therapies are administered can affect progression-free survival. For example, the results of the PROREPAIR-B study (2013) demonstrated that administration of a taxane prior to an androgen signaling inhibitor led to inferior progression-free survival compared with non-carriers of BRCA2\textsuperscript{145}. However, reversing the order of administration led to a similar progression-free survival to non-carriers of the BRCA2 mutation\textsuperscript{145}. Therefore, studies such as these suggest that genetic testing could be used to optimise treatment strategies for prostate cancer patients\textsuperscript{145}.

Phosphodiesterase-4D7 (PDE4D7) is an emerging prognostic biomarker for prostate cancer\textsuperscript{146}. It was recently validated in a 503-patient study, which followed patients for 10 to 15 years to confirm the prognostic, and incremental value of PDE4D7 compared with established clinical risk metrics\textsuperscript{146}. In 2018 MDxHealth Inc announced that it signed a worldwide licensing agreement with Philips for the rights to manufacture and market its tissue-based InformMDx test based on the PDE4D7 biomarker to stratify patients according to their risk of disease progression and the development of secondary tumors\textsuperscript{146}. It is anticipated that this will provide useful information for clinicians to guide post-biopsy treatment decisions at the time of diagnosis, as well as treatment decisions following prostatectomy\textsuperscript{146}.

The main biomarkers used in prostate cancer, the tests used to identify them, and the use of this information in therapy are summarised in Table 6. Despite the gradual introduction of these PM approaches, the mainstay of treatment depending on the stage of the disease is presently based on androgen deprivation therapy, using agents such as luteinising hormone releasing hormone (LHRH) agonists (e.g. luserelin, goserelin, leuprorelin acetate or triptorelin), gonadotrophin-releasing hormone (GnRH) antagonists (e.g. degarelix), anti-androgen small molecule agents (e.g. bicalutamide, flutamide or cyproterone acetate) or traditional chemotherapy (e.g. docetaxel). Surgery, brachytherapy or radiotherapy are also important treatment strategies.

See Supplementary PDF for Table 6: Biomarkers important in prostate cancer, the therapeutic agents associated with them and the biomarker tests used.

Source:\textsuperscript{147}

Renal cell carcinoma
Von Hippel-Lindau (VHL) mutations are associated with many renal cell carcinoma (RCC) tumours resulting in higher levels of the heterodimeric transcription factor HIF1α (hypoxia inducible factor 1-α) and also VEGF (vascular endothelial growth Factor). VHL is associated with an autosomal dominant hereditary disorder, as well as being considered a tumour suppressor. The VHL protein targets the α-subunit of HIF1α and also associates with an E3 ligase, and has an important role in regulating hypoxia pathways both in tumour and healthy tissues,[148][149], and growth factor-β signaling pathways in RCC[150]. Stabilisation of HIF1α corresponds to elevated VEGF levels together with increased vascularisation of VHL-associated tumours[148][149]. A growing knowledge of the importance of VHL-HIF pathway in RCC has led to the development of treatment strategies involving multi-targeted tyrosine kinase inhibitors (e.g. pazopanib) and mTOR inhibitors (e.g. temsirolimus)[149].

VEGF is a potent promoter of tumour angiogenesis, and both mRNA and protein levels of VEGF are higher in RCC cells than in normal kidney cortex cells. In addition, VEGF and PDGF-B (platelet-derived growth factor B) expression have been found to be higher in papillary carcinoma cells than in other RCC subtypes[151]. Although not currently available in the NHS, the NexCourse Complete/Solid Genoptix-A Test has been used experimentally to measure over-expression of VEGFR 1-3 to select patients who may benefit from treatment with pazopanib (see Table 7).

Some patients with RCC have been known to experience innate immune responses to their disease, which has led to exploration of the use of immunotherapies[152]. For example, the PD-L1 inhibitors nivolumab (Opdivo; Bristol-Myers Squibb Pharmaceuticals Ltc) and pembrolizumab (Keytruda; MSD) that inhibit the interaction between PD-L1 and PD-1 have been studied, but no CDx tests are presently available for these agents. Based on clinical studies, Weinstock et al have demonstrated that the PD-1/PD-L1 interaction is an important regulator of tumour immune tolerance and growth in RCC[153], and that by blocking this interaction, progression of the disease can be slowed.

See Supplementary PDF for Table 7: Biomarkers important in renal cancer, the PM agents used in this disease and the companion diagnostic tests used to select patients most likely to respond

Hepatocellular carcinoma

VEGF is a molecular biomarker that participates in processes such as the recruitment of endothelial cells through activation of the SAPK2/p38 MAP kinase module[153], and the activation of receptors involved in the proliferation of tumour cells[154]. Studies have suggested that VEGF has an important role in the development, growth and metastasis of not just HCC, but in a number of other tumours types including lung, gastric, colorectal, and head and neck squamous cancers[154]. For HCC, the tyrosine kinase inhibitor sorafenib was developed to block synthesis of intercellular factors such as VEGF to regulate angiogenesis and thus the progression of the disease[154].

PD-L1 is a co-stimulatory ligand that is commonly expressed by HCC cells both in vitro and in vivo[155]. There is evidence to suggest that HCC patients whose tumours express PD-L1 are at higher risk of cancer reoccurrence. Furthermore, PD-L1 is a prognostic factor for tumour vascular invasion and encapsulation of HCC[155]. Overall, over-expression of PD-L1 in HCC correlates with tumour aggressiveness and an enhanced risk of post-operative recurrence[155]. Therefore, PD-L1 can be used in HCC as a biomarker for both prognosis and therapy with immune-oncology checkpoint inhibitors such as nivolumab (Opdivo; Bristol-Myers Squibb)[155].
The biomarkers used in hepatocellular carcinoma, the tests used to identify them, and the use of this information in therapy are summarised in Table 8.

See Supplementary PDF for Table 8: Biomarkers important in hepatocellular carcinoma, the PM agents used in these diseases and the companion diagnostic tests used to select patients most likely to respond

### Papillary and medullary thyroid cancers

VEGF and PDGFR are commonly over-expressed in thyroid cancers\(^{[156]}\). For example, their presence correlates with an increase in vascularity and microvessel density in a papillary thyroid compared with that of a normal thyroid\(^{[156]}\). Cohen *et al.* have reported that VEGF and PDGFR over-expression are closely linked with tumour stage, size, nodal involvement, extra thyroidal invasion, distant metastasis and the risk of papillary thyroid cancer re-occurrence\(^{[148],[156]}\).

By observing the multiple mutations within the *BRAF* gene, it has been established that its activation and, in turn, activation of the *RAF/MEK/MAPK* signaling pathway, is an important event in the development of papillary thyroid cancer\(^{[157]}\). The presence of the *BRAF*-\(^V600\) mutation was found to be more prevalent in older age groups with lymph node and distant metastases, thus establishing it as an independent prognostic biomarker for recurrent and persistent disease\(^{[158]}\).

The prognostic and diagnostic biomarkers for medullary thyroid cancer have been recently reviewed\(^{[159]}\). Oncogenic activation of *BRAF* contributes to the pathogenesis of several solid tumour types including hepatocellular carcinoma\(^{[160]}\).

More information about the biomarkers and therapeutic agents associated with papillary and medullary thyroid cancer is summarised in Table 9.

See Supplementary PDF for Table 9: Biomarkers used in medullary and papillary thyroid cancers, the tests used to identify them and the use of this information in therapy

### Pancreatic neuroendocrine tumours

Biopsy samples taken from pancreatic neuroendocrine tumours and pancreatic carcinomas frequently show over-expression of multiple biomarkers including PDGFR, C-kit (stem cell factor receptor) and VEGFR\(^{[161]}\). The kinase inhibitor sunitinib (Sutent; Pfizer) is capable of inhibiting these, and can delay the growth of pancreatic islet cell tumours by reducing endothelial cell density and the pericyte coverage of tumour vessels\(^{[161]}\).

ERCC1 (excision repair cross-complementing gene-1) produces an excision nuclease within the nucleotide excision repair pathway. Based on IHC, over-expression of this gene has been shown to be a good predictive biomarker to guide initial treatment\(^{[162]}\). Research has suggested that over-expression of ERCC1 might be an effective predictor of response to FOLFIRINOX (a combination of folinic acid, fluorouracil, irinotecan and oxaliplatin) in metastatic pancreatic cancer\(^{[163]}\).
An emerging area of interest is the identification of pancreatic tumours harbouring \textit{BRCA1/2} mutations that might be treated with PARP inhibitors such as olaparib (Lynparza) targeted to these mutations and other DNA damage repair defects. One study involving 3,315 metastatic pancreatic cancer patients (the POLO trial) utilised the Myriad Genetics BRACAnalysis CDx assay to identify suitable patients, and provided olaparib (Lynparza) to \textit{BRCA1/2}-positive individuals following 16 or more weeks of platinum-based chemotherapy. This approach extended progression-free survival from 3.8 to 7.4 months compared with a placebo trial arm, although no significant health-related quality of life differences between the maintenance treatment and placebo arms was observed\cite{163}. The investigators concluded that a strategic approach of first-line platinum-based chemotherapy followed by maintenance olaparib treatment could become a new standard of care for metastatic pancreatic cancer patients with germline \textit{BRCA1/2} mutations\cite{163}.

The biomarkers used in pancreatic neuroendocrine tumours the tests used to identify them, and the use of this information in therapy is summarised in Table 10.

Bladder cancer

The checkpoint immune-oncology target PD-L1 has been a primary focus for the treatment of bladder cancer\cite{165}, and it has been demonstrated that tumours expressing higher levels of PD-L1 are more likely to be assessed as high-grade, with greater frequencies of post-operative recurrence and poorer survival\cite{165}.

Durvalumab (Imfinzi; AstraZeneca), a monoclonal antibody targeted to PD-L1, was granted ‘breakthrough therapy’ designation by the FDA in 2016 for patients with PD-L1 positive inoperable or metastatic urothelial bladder cancer whose tumours have progressed during or after a standard platinum-based regimen\cite{165}. This new agent is also being studied for its use in the treatment of NSCLC, hepatocellular carcinoma, mesothelioma, and head and neck, pancreatic and haematologic cancers\cite{165}. Atezolizumab (Tecentriq) and Avelumab (Bavencio) are also being studied for use in this disease setting.

A new biomarker gaining relevance to the treatment of bladder cancer is the fibroblast growth factor receptor 2/3 (i.e. FGFR2 and FGFR3)\cite{166}. In 2019, the FDA provided accelerated approval for erdafitinib (Balversa; Janssen Pharmaceuticals), which is targeted, to advanced adult bladder cancer patients with FGFR2 or FGFR3 mutations following treatment with platinum-based agents\cite{166}. The FDA simultaneously approved the Therascreen FGFR RGQ RT-PCR Kit (Qiagen Inc) as a companion diagnostic test to select patients with FGFR3 or FGFR2 mutations who should benefit from erdafitinib therapy\cite{166}. This was the first FDA approval of an FGFR-related therapeutic agent or companion diagnostic test\cite{166}. 

See Supplementary PDF for Table 10: Biomarkers used in pancreatic neuroendocrine tumours (PNTs), the tests used to identify them and the use of this information for therapy
As bladder tumours progress, they tend to spread into the muscularis propria surrounding the bladder. It has been shown that increased mutational frequency in genes that encode proteins involved in chromatin remodeling may prove useful in predicting whether a non-invasive tumour progresses to muscle invasion\textsuperscript{[167]}. Two subtypes of non-invasive bladder cancer, GS1 and GS2, have been classified based on their genomic signatures, and in particular on an increase in Ki67 labelling and up-regulation of mTORC1 signaling\textsuperscript{[168]}. Interestingly, mutations in the histone demethylase enzyme-coding gene KDM6A are also thought to play a role, but are found predominantly in female patients\textsuperscript{[167]}. These diagnostic and prognostic biomarkers appear to be key oncogenic drivers of the disease, and may prove to be useful therapeutic targets in the future\textsuperscript{[166],[167]}.

The main biomarker used in bladder cancer, the test used to identify it, and the use of this information in therapy is summarised in Table 11.

See Supplementary PDF for Table 11: The main biomarker used in bladder cancer, the test used to identify it and the use of this information in therapy

### Basal cell carcinoma

Basal cell carcinoma (BCC) is the most common type of non-melanoma skin cancer\textsuperscript{[169]}. It is rarely treated with targeted therapeutic agents, the main treatment strategies being surgery, topical cytotoxic agents (e.g. 5-fluorouracil) and/or radiotherapy\textsuperscript{[168]}. The occurrence of metastasis for BCC is very low (i.e. 0.0028–0.5500%) and, if diagnosed early, surgery is usually curative\textsuperscript{[168]}. However, there has been an effort to develop targeted agents for patients in whom a BCC has recurred following surgery or radiation therapy, or who are not candidates for surgery or radiation therapy\textsuperscript{[170]}.

Hedgehog signaling is an important component of normal cellular development, regulating key target genes involved in modulation of the microenvironment (see Figure 3\textsuperscript{[171],[172]}). This pathway involves binding of the ligand SHH (‘sonic hedgehog’) to the PTCH1 (‘patched-1’) receptor\textsuperscript{[171]}. In the absence of ligand, PTCH1 inhibits SMO (smoothened), a downstream protein in the pathway whose role is to migrate from the intracellular endosome to the cell membrane of the cilium where it activates the glioma-associated oncogene that stimulates expression of target genes\textsuperscript{[170]}. 
In oncogenesis, hedgehog signalling can influence tumour cell growth, differentiation and immune regulation, thus promoting disease progression and metastasis\[^{171}\]. BCC and medulloblastomas are the two most common cancers known to be associated with mutations in components of the hedgehog signalling pathway\[^{172}\]. In 2012, the FDA approved the orally administered hedgehog inhibitor vismodegib (Erivedge; Roche) for use in the treatment of BCC\[^{173}\]. This agent works by suppressing the hedgehog signalling pathway by binding to SMO (see Figure 3), thus inhibiting down-stream activation of the glioma-associated oncogene\[^{172}\]. It is a useful adjunct to therapy in cases where complete surgical resection is not achievable\[^{172}\].

Sonidegib (Odomzo; Novartis) is a second orally available analogue\[^{171}\]. It also binds to and inhibits SMO to prevent activation of the hedgehog pathway\[^{171}\]. The FDA approved it in 2015 for treating patients with locally advanced BCC that has recurred following surgery or radiation therapy, or for those who are not candidates for surgery or radiation therapy\[^{171}\]. There are currently no CDx tests available to evaluate the status of the hedgehog signalling pathway.

The main biomarker relevant to basal cell carcinoma, and the small molecule inhibitors associated with it are summarised in Table 12.
Gastrointestinal stromal tumours

GISTs most commonly arise within the submucosa of the stomach and small bowel, with the cells of origin thought to be the pacemaker interstitial cells of Cajal. Diagnosis is supported with a positive immunohistochemistry for DOG1 (a calcium-dependent receptor-activated chloride channel) or C-Kit (also known as CD117) (see Table 13)[173][174].

The majority of GISTs (i.e. around 85 to 90% of cases) have activating mutations in the C-Kit or PDGFRA receptor tyrosine kinases (TKIs). The C-kit mutations occur most frequently within exon 11, and affected tumours demonstrate a superior response to TKIs. The response of tumours with exon 9 mutations is less favourable, and those tumours with exon 17 mutations are usually resistant to first generation TKIs. Mutations in PDGFRA most commonly occur in exon 18, and affected tumours are also resistant to first generation TKIs[173][174][175].

Treatment with the first generation TKI imatinib (Glivec, Novartis) for three years is recommended as first-line adjuvant therapy for GISTs when there is a high risk of relapse owing to incomplete surgical excision or metastatic disease[173][176]. Sunitinib (Sutent, Pfizer) is given when there are intolerable side effects with imatinib or tumour resistance develops[177]. Regorafenib (Stivarga, Bayer) is a second-generation multikinase inhibitor with FDA approval as an alternative therapy when there is intolerance to the other TKIs, or tumour resistance develops with both imatinib and sunitinib[174][178].

Soft-tissue sarcomas

While rare, soft-tissue sarcomas (STSs) have an increasing incidence, although this currently remains under 1% of the UK population[180]. This heterogeneous group of malignant mesenchymal tumours has more than 50 subtypes, traditionally classified according to tissue differentiation that arise from a range of anatomical sites and demonstrate diverse clinical behaviour[181]. While the primary treatment is surgical resection or radiotherapy-based if resection is unachievable, tumour sensitivity to cytotoxic chemotherapies is also recognised in some subtypes[182][183]. An understanding of the molecular biology of these tumours has led to the identification of diagnostic genetic aberrations. Around 20% of STS subtypes demonstrate an identifiable chromosomal translocation, of which around 66% result in fusion-protein activated pathways to which therapies may be targeted[184]. STS translocation aberrations and associated targets have been categorised according to fusion gene protein product function as previously reviewed along with suitable targeted therapeutic approaches[181][184][182][183][185][186].
For example, PAX3-FOXO1 and PAX7-FOXO1 gene fusions are recurrently associated with morphologically distinctive alveolar rhabdomyosarcoma (ARMS), and are identifiable in 65% and 20% of cases respectively (184). ARMS most commonly presents in the group aged under 15 years, and the presence of a FOXO1 translocation is associated with a poorer prognosis. However, it has proven difficult to target chimeric transcription factor proteins directly (183). Similarly, Ewing sarcoma expresses prototypical transcription factor gene fusions such as EWSR1-FLI1 that is associated with down-stream over-expression of IGF1R (insulin growth factor 1 receptor) (185). While Ewing sarcoma is typically chemotherapy sensitive (i.e. 70% of patients have five-year event-free survival in the non-metastatic group), recurrence is a poor prognostic scenario in which partial tumour response to the IGFR1 inhibitor trabectedin (Yondelis, Immedica) has been demonstrated (186). Trabectedin has European (2007) and FDA (2015) approval for the treatment of metastatic liposarcoma and leiomyosarcoma, with other IGFR1 inhibitors such as figitumumab and ganitumab providing objective responses as mono- or combination-therapies in early- and late-phase STS clinical trials respectively (183).

Other recurrent gene fusions occur in the protein kinases (184), and TKIs have provided some overall survival benefit in subsets of STSs. For example, activity of the VEGF inhibitor pazopanib in non-adipocytic STS contributed toward FDA approval in 2012 for second-line use in this disease (182). Sunitinib also has activity against VEGFR, PDGFR and RET, and cediranib has activity against multiple VEGFRs and KIT. Both of these TKIs are showing promise for the treatment of alveolar soft-part sarcomas in early-phase clinical trials (183). In addition, mTOR inhibitors acting down-stream of PI3K/AKT in combination with TKIs or IGFR1 inhibitors have provided partial responses, but this is typically limited to a few individual STS patients in which general toxicity remains a major limitation (183). Therefore, moving forward, there is a need to utilise biomarkers for the stratification of STS patients into those who are most likely to benefit from the increasing range of therapies available (183).

Tumour agnostic agents and related biomarkers

Until recently, novel PM agents were designed (and approved) against cancer types originating in specific tissues within the body (e.g. breast, bowel and lung) based on particular genetic aberrations in biopsied cells (e.g. vemurafenib for BRAF\textsuperscript{V600E} mutations in melanoma). However, there is a current trend to develop novel PM agents targeted towards biomarkers irrespective of which tumour or tissue types they arise in (i.e. so-called ‘tissue agnostic’ or ‘pan anticancer’ agents) (187). (188).

For example, in 2014 the FDA initially approved pembrolizumab (Keytruda, MSD) under the FDA Fast Track Development Program for use following treatment with ipilimumab (or after treatment with ipilimumab and a BRAF inhibitor) in advanced melanoma patients carrying a BRAF mutation (189). However, in 2017 it was approved for use in non-small cell lung cancer, recurrent or metastatic head and neck cancer, refractory classical Hodgkin lymphoma, and urothelial carcinoma (190). This agent works by targeting the cellular pathway involving PD-1/PD-L1, a receptor and ligand found on T-cells and some cancer cells respectively (191). By blocking this pathway, pembrolizumab stimulates the body’s immune system to kill the tumour cells.
Then, in 2017, the FDA granted accelerated approval for the use of pembrolizumab in the treatment of any adult or paediatric patients whose cancers have the specific genetic biomarkers of microsatellite instability-high (MSI-H) or mismatch repair deficiency (dMMR)\textsuperscript{[114],[192]}. This was the first time that the FDA had approved a cancer treatment based on common biomarkers without regard for the tumour's original location in the body\textsuperscript{[115],[189]}. MSI-H and dMMR are genetic abnormalities that, if present, reduce the ability of cells to repair their DNA after mutation or other damage\textsuperscript{[193]}. Tumours with these biomarkers are most commonly found in colorectal, endometrial and gastrointestinal cancers, but are also less commonly found in breast, prostate, bladder and thyroid cancers\textsuperscript{[113],[194]}. Crucially, around 5% of patients with mCRC have MSI-H or dMMR biomarkers in their tumours\textsuperscript{[187],[193],[195]}. Pembrolizumab was approved for this new indication using the accelerated approval pathway, through which the FDA may approve drugs for serious conditions where there is an unmet clinical need\textsuperscript{[114]}.

The safety and efficacy of pembrolizumab for this new approval were studied in patients carrying MSI-H or dMMR in their solid tumours enrolled in one of five uncontrolled, single-arm clinical trials. In some trials, patients were required to already have MSI-H or dMMR cancers prior to treatment, while in others a subgroup of patients were identified as having MSI-H or dMMR cancers by testing tumour samples after treatment had started. A total of 15 cancer types were identified among 149 patients enrolled across these five clinical trials, the most common being colorectal, endometrial and other gastrointestinal cancers. Of the 149 patients who received pembrolizumab in these trials, 39.6% had a complete or partial response, and for 78% of these patients, the response lasted six months or more\textsuperscript{[196]}.

In another example, researchers at Array BioPharma discovered larotrectinib (Vitrakvi) (see Figure 4), an inhibitor of the Tropomyosin Kinase Receptors TrkA, TrkB and TrkC, which was subsequently licensed to Bayer and Loxo Oncology\textsuperscript{[196]}. Larotrectinib was initially awarded orphan drug status by the FDA in 2015 for soft tissue sarcoma, which was followed by breakthrough therapy designation in 2016 for treatment of the approximately 1% of patients whose metastatic tumours are caused by chromosomal TRK gene fusions\textsuperscript{[196],[197],[198]}. Clinical trial results were announced in 2017 and, as a result, the FDA approved larotrectinib in November 2018\textsuperscript{[199]}.
Larotrectinib (Vitrakvi) is the first small molecule agent to be approved to treat any cancers containing certain mutations, rather than cancers of specific tissues (i.e. the approval was ‘tissue agnostic’).

Larotrectinib was the first small molecule agent to be specifically developed and approved to treat any cancers containing certain mutations, rather than cancers of specific tissues (i.e. the approval was ‘tissue agnostic’). The fact that larotrectinib reduced tumour size in 76% of patients carrying the TRK fusion genes across many types of cancers, prompted Bayer to pay US$1.5bn for the rights to jointly develop it, along with a follow-on agent Loxo-195. Although the annual cost of treatment with larotrectinib is expensive (i.e. around US$393,600), the TRK fusion is so rare that oncologists do not routinely look for them, which partly explains the high price tag aimed at recovering research costs and the relatively low sales volume anticipated.

Based on these two promising examples of pembrolizumab (Keytruda) and larotrectinib (Vitrakvi), it is likely that more tissue agnostic agents will be produced and approved in the future, which should provide greater opportunities to treat advanced cancer patients regardless of their tumor type and, in particular, will enable new treatment opportunities for those with rare or hard to treat diseases. This should encourage oncologists to consider the molecular abnormalities that may be driving their patients’ tumours at the outset, and not just the organs affected.

Supportive therapies

PMs that target tumour cells while leaving healthy cells intact should, in principle, be devoid of side effects. Although this goal may be reached in the future, at present most, if not all, of the anticancer agents used in the clinic, whether classed as PMs or not, produce side effects in patients. Therefore, a number of supportive medicines are usually provided to treat these side effects to make the anticancer therapies more tolerable. They are essential for maintaining a good quality of life during treatment schedules, as well as ensuring that a patient completes their treatment without delays or dose reductions. These supportive therapies are usually tailored to individual patients depending on which chemotherapy regimen they are receiving and which side effects they encounter, so in this context they are being used as part of a PM approach.

Examples of supportive therapies include anti-emetics (e.g. ondansetron) to treat nausea and vomiting, and granulocyte-colony stimulating factors (GCSF) to reduce the risk of serious infection from myelosuppression. The majority of patients will be given anti-emetics before they receive chemotherapy and on the days following treatments, and a number of different agents may be tried before the best one for a particular patient is identified. If a PM agent is being used in combination with traditional anticancer regimens, other examples of supportive therapies include pyridoxine to counter the palmar-plantar erythrodysesthesia resulting from fluorouracil or capecitabine treatment, folic acid to reduce the toxicity of pemetrexed or methotrexate, and omeprazole to protect the stomach from steroid-based treatments.

The other major supportive medication is opioid therapy for pain associated with a tumour impacting on bones, nerves or other organs. Numerous opioid-based medications are available, and adjustments are made for individual patients to ensure that the best agent, dose and/or formulation are being used.

Funding of cancer drugs within the NHS
The growing number of new cancer therapies available, and their relatively high cost, places an increasing financial burden on the NHS and other healthcare systems globally. The new generations of PMs are particularly expensive owing to development costs and the fact that drug companies will sell fewer units owing to patients being selected for treatment (i.e. unlike the previous ‘block-buster’ model of drug sales where high demand reduced unit prices). Also, the use of PMs can be more expensive due to the need for CDx tests. Most observers agree that current funding models within the NHS are unsustainable, and that a major overhaul of how the system provides expensive anticancer agents to the entire UK population is required.

At present, the provision of cancer drugs in England is a complicated process with multiple streams of funding. A number of anticancer drugs have been approved by NICE and are available through NHS England. In addition, a small number of anticancer medications are funded locally through clinical commissioning groups. In April 2011, the Cancer Drugs Fund (CDF) was introduced as a temporary solution to ensure that high-cost anticancer agents not routinely available through the NHS could be made available to patients. However, there was little control around the criteria for supplying drugs through the CDF, and the timeline for when drugs should exit the fund was not clear, which resulted in an increasing spend each year. It was agreed that the CDF was not a sustainable method for providing drugs to patients, and so a public consultation was conducted in 2015 by NHS England to establish a new model, with NICE overseeing the evaluation process, which was introduced in July 2016.

In this new model, any new anticancer agents that are expected to receive marketing authorisation, or proposals for new indications for existing agents, are now referred to NICE for an initial appraisal. The advantage is that, as well as containing a clear exit strategy, the budget will not be allowed to overspend. However, if it does, the originating pharmaceutical company has a responsibility to contribute financially back into the CDF for their particular drug. At the point of appraisal by NICE, there are three potential outcomes. First, the new agent or new indication can be routinely commissioned, meaning that it will be funded by the CDF for 90 days. Following publication of the final NICE technology appraisal, it is then funded by NHS England. Second, the agent or new indication can be denied approval, meaning that it will only be available in exceptional circumstances through an individual funding request. Third, the new agent or indication can be entered into the interim CDF under a ‘managed access scheme’ to allow for more data to be collected. This option allows new agents or indications to be funded for a maximum of two years via the CDF, at which point a decision is made as to whether it will be routinely commissioned or not. Alternative access schemes can be either funded by the originating pharmaceutical company (i.e. through ‘compassionate access schemes’) or are evaluated via the MHRA through ‘early access to medicines’ schemes. Although these schemes are useful for providing new treatments to patients, they require a significant amount of administration and management resources.

Funding of biomarker assays for cancer therapies

Although new PM therapies themselves will represent a significant cost to the NHS, the pharmacogenomic tests (i.e. CDxs) needed to support them will also add a significant cost. Most of the examples provided in this review are typically single biomarker assays, with a few having the potential to be carried out in parallel within the same assay (a process known as multiplexing). Eventually, multiplexing could reduce costs through, for example panel testing or whole exome sequencing.
Overall, advances in NGS technologies have greatly enhanced the development and use of biomarker-driven cancer therapies. Genomic profiling has become significantly more cost-effective since the Human Genome Project, when the cost of sequencing the human genome was many millions of pounds and took more than a decade to complete. Current NGS platforms can sequence a patient's genome in a few days at a cost of around £1,000. In particular, the affordability of, and improvements to, NGS have led to the commercialisation of NGS platforms that now have widespread use in both research and clinical decision-making.

Despite the greater availability of tumour molecular profiling by NGS, the goal of improving patient outcomes while reducing overall costs (sometimes known as 'value-based care') remains challenging. A recent review by Gong et al. has discussed the financial modelling of cost (efficiency) versus clinical benefit (effectiveness) and toxicity (safety) in relation to genomic profiling for cancer care, and it is clear that many health regimens around the world will find it challenging to deliver state-of-the-art genomics-based cancer care to all patients. However, it is predicted that in the near future advances in NGS techniques with high-throughput functionality will enable the massive parallel sequencing of genomes at unprecedented rates (with targeted panels based on whole-exome or whole-genome sequencing), which should lower costs significantly.

Conclusion

In the past three decades, the PM approach to cancer therapy has progressed from the concept phase to practical utility. This has significant implications for healthcare systems, such as the NHS, in that it creates the opportunity to provide anticancer agents to patients with a high certainty of providing clinical benefit. However, these new therapies are expensive and require CDx tests to select patients. Although pharmaceutical companies are adjusting to this new paradigm by setting higher prices for novel PM-based anticancer therapies that sell in lower volume, government-funded healthcare organisations such as the NHS will come under increasing pressure to fund these new therapies because of their clinical effectiveness.

For these new PM approaches, the growing burden on clinicians, pharmacists and other healthcare professionals will become increasingly significant due to the additional requirements for identifying suitable patients using diagnostic tests followed by appropriate drug selection and variable dosing based on the test results. The complexity of this new treatment paradigm demands that healthcare professionals have not only the technical expertise and knowledge to deliver it, but also have the necessary communication skills to explain diagnostic test results and drug selection decisions clearly and confidently to patients and their carers, and other healthcare professionals.

This review has provided an overview of the current status of the PM approach to cancer therapy with regard to the main types of solid tumours. A second part of this review, focusing on haematological malignancies, will also include a summary of the potential future applications of biomarkers in oncology.

It should be noted that the accuracy and coverage of this review (and the one following on haematological malignancies) will be short-lived owing to the rapid advances being made in this area, with new PM therapies and biomarker assays entering the clinic at a remarkable rate. Each section of this review represents only a brief summary of what ideally should be a systematic review of thousands of publications. The reader should be aware of this, and is encouraged to look further into the literature in their area of interest.

Disclaimer
The treatment strategies described in this review are for educational purposes only and should not be used to guide the treatment of patients. Readers are referred to NICE or SIGN guidance in the UK, and relevant medical texts and specialist journals, for information about prescribing and treatment regimens. The authors alone are responsible for the views expressed in this article, which do not necessarily represent the views, decisions or policies of the institutions with which they are affiliated.

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Table 1: Biomarkers important in breast cancer, the precision medicine (PM) agents used in this disease, and the companion diagnostic (CDx) tests used to select patients most likely to respond

<table>
<thead>
<tr>
<th>Biomarkers</th>
<th>Agents used</th>
<th>Examples of companion diagnostic tests</th>
<th>Use of test results</th>
</tr>
</thead>
<tbody>
<tr>
<td>ER</td>
<td>Anastrozole (Arimidex)</td>
<td>Immunohistochemistry(^{\text{NHS}}) (IHC)</td>
<td>IHC is assessed as intensity (1–3) and proportion of cells stained (0–5) to provide a score of 0–8 (known as the Allred score)</td>
</tr>
<tr>
<td></td>
<td>Exemestane (Aromasin)</td>
<td>EndoPredict gene expression profiling assay (Myriad Genetics)(^{\text{nNHS}})</td>
<td>Identifies ER+ve tumours. Not cost effective for NHS</td>
</tr>
<tr>
<td></td>
<td>Everolimus (Afinitor)</td>
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<td></td>
<td>Letrozole (Femara)</td>
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<td></td>
<td>Fulvestrant (Faslodex)</td>
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<tr>
<td></td>
<td>Tamoxifen (Nolvadex)</td>
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<td></td>
<td>Palbociclib (Ibrance)</td>
<td></td>
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<tr>
<td></td>
<td>Ribociclib (Kisqali)</td>
<td></td>
<td></td>
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<tr>
<td></td>
<td>Raloxifene Hydrochloride* (Evista)</td>
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<tr>
<td></td>
<td>Toremifene* (Fareston)</td>
<td></td>
<td></td>
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<tr>
<td>PgR</td>
<td>Everolimus (Afinitor)</td>
<td>EndoPredict Gene Expression Profiling Assay (Myriad Genetics)(^{\text{nNHS}})</td>
<td>Identifies PgR+ve tumours. Not considered to be cost effective for NHS</td>
</tr>
<tr>
<td></td>
<td>Toremifene* (Fareston)</td>
<td>Oncotype DX (Genomic Health Inc)(^{\text{nNHS}})</td>
<td>Assesses whether a patient is likely to benefit from chemotherapy(^{\text{66}}). Not considered to be cost effective for NHS</td>
</tr>
<tr>
<td>HER2/neu</td>
<td>Lapatinib*** (Tyverb)</td>
<td>Immunohistochemistry\textsuperscript{NHS}</td>
<td>Score of 3+ defined as positive, 2+ equivocal</td>
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<td></td>
<td></td>
<td>HER2 IQFISH PharmDX assay\textsuperscript{NHS} (Dako Omnis)</td>
<td>Used for direct confirmation of HER2 status, for example to check for amplification of HER2+ antigen in breast tumour cells after an equivocal HER2 IHC result of 2+</td>
</tr>
<tr>
<td></td>
<td></td>
<td>HercepTest (Dako PharmDx) — a semiquantitative IHC assay\textsuperscript{NHS}</td>
<td>Used to select patients for treatment with trastuzumab and pertuzumab</td>
</tr>
<tr>
<td></td>
<td>Trastuzumab (Herceptin)</td>
<td>Trastuzumab Emtansine (Kadcyla)</td>
<td>Pertuzumab (Perjeta)</td>
</tr>
<tr>
<td>Mutated \textit{BRCA1} and \textit{BRCA2}</td>
<td>Olaparib* (Lynparza)</td>
<td>BRCA Analysis CDx (Myriad Genetics Inc)\textsuperscript{NHS}</td>
<td>Identifies ovarian cancer patients carrying germline \textit{BRCA} mutations who may benefit from treatment with olaparib or talazoparib\textsuperscript{70}</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Talazoparib** (BMN-673)</td>
<td></td>
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<tr>
<td>CDK4 and CDK6</td>
<td>Abemaciclib (Verzenio)</td>
<td>None</td>
<td>-</td>
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<tr>
<td></td>
<td>Palbociclib (Ibrance)</td>
<td>Ribociclib (Kisqali)</td>
<td></td>
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<tr>
<td>PIK3CA</td>
<td>Alpelisib** (Piqray)</td>
<td>Qiagen’s Therascreen PIK3CA RGQ PCR Kit</td>
<td>Used to detect mutations in PIK3CA found in breast cancer patients</td>
</tr>
</tbody>
</table>

\* agents that are likely to be approved by National Institute for Health and Care Excellence (NICE) in the near future, but may not necessarily be used in the NHS\textsuperscript{40,71}  
\** agents that are unlicensed in the UK at the time of writing  
\*** agents that have been removed from NICE guidance or rejected by NICE  
\textsuperscript{NHS} companion diagnostic tests which are used in the NHS based on cost/benefit ratio\textsuperscript{72,73}  
\textsuperscript{nNHS} companion diagnostic tests which are not used in the NHS based on cost/benefit ratio\textsuperscript{72,73}
<table>
<thead>
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<th>Use of test results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Epidermal growth factor receptor tyrosine kinase (EGFR-TK) EGFR1/Human epidermal growth factor receptor 1 (HER1)</td>
<td>Gefitinib (Iressa)† Erlotinib (Tarceva)‡</td>
<td>The Cobas EGFR Mutation Test v2&lt;sup&gt;NHS&lt;/sup&gt; (Roche Molecular Systems, Inc)‡</td>
<td>Used to select patients with non-small cell lung cancer (NSCLC) who may benefit from gefitinib and erlotinib</td>
</tr>
<tr>
<td>EGFR/Erb-B family</td>
<td>Necitumumab*** (Portrazza)</td>
<td>Thermo Fisher OncomineDx Target Test&lt;sup&gt;NHS&lt;/sup&gt; (Thermo Fisher Scientific)</td>
<td>OncomineDx Target is a 23-gene test used to identify the best responders to agents working through the genes identified (i.e. EGFR, BRAF, ALK, ROS-1, KRAS and NRAS). It can also be used to monitor the presence or absence of variants in other genes&lt;sup&gt;91–93&lt;/sup&gt;</td>
</tr>
<tr>
<td>EGFR (sensitivity toward T790M mutation) and with Exon 19 deletions or Exon 21 L858R mutations</td>
<td>Afatinib (Giotrif)§ Dacomitinib (Vizimpro)</td>
<td>EGFR Pharm Dx test&lt;sup&gt;NHS&lt;/sup&gt; (Dako Inc) and the Therascreen EGFR RGQ PCR test&lt;sup&gt;NHS,94&lt;/sup&gt;</td>
<td>Both EGFR tests are used to guide the selection of patients who may benefit from treatment with afatinib</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Therascreen EGFR RGQ PCR Kit&lt;sup&gt;NHS,95&lt;/sup&gt;</td>
<td>This test detects exon deletions and insertions in the EGFR gene&lt;sup&gt;95&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td></td>
<td>The Cobas EGFR V2 test&lt;sup&gt;NHS&lt;/sup&gt; (Roche Molecular Systems, Inc)</td>
<td>The V2 test is used to select NSCLC patients who may benefit from treatment with osimertinib</td>
</tr>
<tr>
<td>EGFR and HER2 (Exon 20 insertion mutation in EGFR)</td>
<td>Poziotinib** (also known as NOV120101 or HM781-36B)&lt;sup&gt;96&lt;/sup&gt;</td>
<td>Thermo Fisher OncomineDx Target Test&lt;sup&gt;NHS&lt;/sup&gt; (Thermo Fisher Scientific)</td>
<td>OncomineDx Target is a 23-gene test used to identify best responders to agents working through the genes identified (i.e. EGFR, BRAF, ALK, ROS-1, KRAS and NRAS). It can also be used to monitor the presence or absence of variants of other genes&lt;sup&gt;91–93&lt;/sup&gt;</td>
</tr>
<tr>
<td>Anaplastic lymphoma kinase (ALK)</td>
<td>Ceritinib (Zykadia)</td>
<td>Ventana ALK (D5F3) Assay&lt;sup&gt;NHS&lt;/sup&gt; (Novartis in collaboration with Roche)</td>
<td>Results from this test at the time of diagnosis can help determine whether patients may benefit from treatment with ceritinib and/or alectinib</td>
</tr>
<tr>
<td></td>
<td>Alectinib (Alecensa)</td>
<td>HTG EdgeSeq ALKPlus Assay EU&lt;sub&gt;NHS&lt;/sub&gt; (HTG Molecular Diagnostics)</td>
<td>HTG EdgeSeq ALKPlus is an &lt;em&gt;in vitro&lt;/em&gt; next-generation sequencing assay used to test for ALK status in NSCLC patients. It works in conjunction with the HTG EdgeSeq Analyser&lt;sup&gt;97&lt;/sup&gt;</td>
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<td>---</td>
</tr>
<tr>
<td></td>
<td>Brigatinib (Alunbrig)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Crizotinib (Xalkori)</td>
<td>Thermo Fisher OncomineDx Target Test&lt;sup&gt;NHS&lt;/sup&gt; (Thermo Fisher Scientific)</td>
<td>OncomineDx Target is a 23-gene test that can be used to identify potential best responders to crizotinib, and is also used to monitor for the presence or absence of variants of other genes&lt;sup&gt;91–93&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>ROS-1</td>
<td>Lorlatinib (PF-6463922)</td>
<td>This test is used only to detect the mutation and not for drug selection</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Vascular endothelial growth factor (VEGFR), platelet-derived growth factor (PDGFR) and fibroblast growth factor (FGFR)</td>
<td>Nintedanib (Vargatef)</td>
<td>NexCourse Complete/Solid Test&lt;sup&gt;NHS&lt;/sup&gt; (Genoptix Inc)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Ramucirumab*** (Cyramza)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>BRAF&lt;sup&gt;V600E/K&lt;/sup&gt;</td>
<td>Dabrafenib*** (Tafinlar)</td>
<td>Thermo Fisher OncomineDx Target Test&lt;sup&gt;NHS&lt;/sup&gt; (Thermo Fisher Scientific)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>OncomineDx Target is a 23-gene test that can be used to identify best responders to the genes identified (i.e. EGFR, BRAF, ALK, ROS-1, KRAS and NRAS), and also to detect the presence or absence of variants of other genes&lt;sup&gt;91–93&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>PD-L1²</td>
<td>Durvalumab (Imfinzi)</td>
<td>Nivolumab (Opdivo)</td>
</tr>
<tr>
<td>---------------</td>
<td>-----------------------</td>
<td>---------------------</td>
<td>--------------------</td>
</tr>
<tr>
<td>MEK</td>
<td>Trametinib*** (Mekinist)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>The Ventana PD-L1 (SP263) Assay NHS (Ventana Medical Systems Inc)</td>
<td>PD-L1 IHC 28-8 PharmDxTest⁴⁸⁸⁸ (Agilent Technologies/Dako Inc)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Used to select patients who may benefit from treatment with durvalumab</td>
<td>Used in the selection of patients who may benefit from treatment with nivolumab</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TMB (not currently used in practice)</td>
<td>Atezolizumab (Tecentriq)</td>
<td>None</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td>Avelumab** (Bavencio)</td>
<td>None</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td>Tramelimumab** (CP-675,206)</td>
<td>None</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td>Ipilimumab** (Yervoy)</td>
<td>FoundationOne CDx⁴⁸⁸⁸ NHS (FoundationFocus Inc)</td>
<td></td>
</tr>
</tbody>
</table>

*agents that are likely to be approved by National Institute for Health and Care Excellence (NICE) in the near

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future, but may not necessarily be used in the NHS\textsuperscript{40,71}  
** agents that are unlicensed in the UK at the time of writing  
*** agents that have been removed from NICE guidance or rejected by NICE  
\textsuperscript{NHS} companion diagnostic tests which are used in the NHS based on cost/benefit ratio\textsuperscript{72,73}  
\textsuperscript{nNHS} companion diagnostic tests which are not used in the NHS based on cost/benefit ratio\textsuperscript{72,73}  
† Both gefitinib and erlotinib are first-generation TKIs  
‡ The Idylla\textsuperscript{TM} EGFR Mutation Assay is used only for research and not for diagnostic purposes  
§ Afatinib is considered a second-generation inhibitor  
Δ Osimertinib is considered a third-generation inhibitor. The decision by NICE to disallow the use of osimertinib for end-of-life consideration in this cancer type is being appealed by AstraZeneca (100)  
◊ The PD-L1 assays described in this section can be used with more than one type of PD-L1 inhibitor  

Table 3: Biomarkers important in metastatic colorectal cancer (mCRC), the precision medicine (PM) agents used in this disease and the companion diagnostic (CDx) tests used to select patients most likely to respond  

<table>
<thead>
<tr>
<th>Biomarkers</th>
<th>Agents used</th>
<th>Examples of companion diagnostic tests</th>
<th>Use of test results</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>NRAS/KRAS</strong></td>
<td>Panitumumab (Vectibix)</td>
<td>Idylla NRAS-BRAF Mutation Test\textsuperscript{NHS} (Biocartis/Amgen)\textsuperscript{116}</td>
<td>Used to help identify mCRC patients eligible for treatment with panitumumab based on a lack of mutations</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Cobas KRAS Mutation Test\textsuperscript{NHS} (Roche Molecular Systems Inc)</td>
<td>Used to help identify mCRC patients who may benefit from treatment with cetuximab or panitumumab based on a lack of mutations</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Therascreen KRAS RGQ PCR Test\textsuperscript{NHS} (Quiagen Ltd)</td>
<td></td>
</tr>
<tr>
<td>HER3</td>
<td>Panitumumab (Vectibix)</td>
<td>Idylla NRAS-BRAF Mutation Test\textsuperscript{NNHS} (Biocartis/Amgen)\textsuperscript{116}</td>
<td>Used to help identify mCRC patients suitable for treatment with panitumumab. Not considered to be cost effective for the NHS</td>
</tr>
<tr>
<td>---</td>
<td>---</td>
<td>---</td>
<td>---</td>
</tr>
<tr>
<td>VEGF-A</td>
<td>Bevacizumab*** (Avastin)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td>VEGFR2</td>
<td>Ramucirumab* (Cyramza)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td>VEGFR2-TIE2</td>
<td>Regorafenib (Stivarga)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td>UGT1A1</td>
<td>Irinotecan (Campto)</td>
<td>UGT1A1 Molecular Assay\textsuperscript{NNHS} (Pfizer)</td>
<td>The presence of the polymorph UGT1A1*28 is associated with poor metabolism of the irinotecan metabolite (SN-38), and the need for a lower starting dose (or avoidance of treatment) due to potentially severe hematological toxicity</td>
</tr>
<tr>
<td>Agents</td>
<td>Pembrolizumab* (Keytruda)</td>
<td>See entry for pembrolizumab in Table 2</td>
<td>See entry for pembrolizumab in Table 2</td>
</tr>
<tr>
<td>--------</td>
<td>--------------------------</td>
<td>----------------------------------------</td>
<td>----------------------------------------</td>
</tr>
<tr>
<td>Nivolumab* (Opdivo)</td>
<td>See entry for nivolumab in Table 2</td>
<td>See entry for nivolumab in Table 2</td>
<td></td>
</tr>
<tr>
<td>Ipilimumab* (Yervoy)</td>
<td>FoundationOne CDx^{NHS} (FoundationFocus Inc)^{98}</td>
<td>The FoundationOne CDx test profiles 324 genes and is used to help identify potential best responders to 15 FDA-approved treatments. Although it is not formally recognised as a companion diagnostic kit for MSI-H and dMMR, it can be used to inform the clinical management of patients with respect to these biomarkers^{87}</td>
<td></td>
</tr>
<tr>
<td>Atezolizumab** (Tecentriq)</td>
<td>None</td>
<td>-</td>
<td></td>
</tr>
</tbody>
</table>

* agents that are likely to be approved by National Institute for Health and Care Excellence (NICE) in the near future, but may not necessarily be used in the NHS^{40,71}

** agents that are unlicensed in the UK at the time of writing

*** agents that have been removed from NICE guidance or rejected by NICE

^{NHS} companion diagnostic tests which are used in the NHS based on cost/benefit ratio^{72,73}

^{nNHS} companion diagnostic tests which are not used in the NHS based on cost/benefit ratio^{72,73}
Table 4: Biomarkers important in metastatic melanoma, the precision medicine (PM) agents used in this disease and the companion diagnostic (CDx) tests used to select patients most likely to respond

<table>
<thead>
<tr>
<th>Biomarkers</th>
<th>Agents used</th>
<th>Examples of companion diagnostic tests</th>
<th>Use of test results</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>BRAF&lt;sup&gt;V600&lt;/sup&gt;</strong></td>
<td>Vemurafenib (Zelboraf)</td>
<td>Cobas 4800 BRAF V600 Mutation Test NHS (Roche Molecular Systems, Inc)</td>
<td>Indicates the presence of the BRAF&lt;sup&gt;V600&lt;/sup&gt; mutation identifying patients suitable for treatment with vemurafenib</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Thermo Fisher Oncomine Dx Target Test&lt;sup&gt;NHS&lt;/sup&gt; (Thermo Fisher Scientific)</td>
<td>Identifies those patients who might best respond to BRAF inhibitors based on the profiling of 23 genes including EGFR, BRAF, ALK, ROS-1, KRAS and NRAS (91-93). Not considered to be cost-effective for the NHS</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Idylla BRAF Mutation Test&lt;sup&gt;NHS&lt;/sup&gt; (Biocartis, Inc)</td>
<td>Detects V600E/E2/D and V600K/R/M mutations in codon 600 of the BRAF gene, and is used to help identify patients who may benefit from BRAF inhibitors&lt;sup&gt;127&lt;/sup&gt;. Not considered to be cost effective for the NHS</td>
</tr>
<tr>
<td><strong>BRAF&lt;sup&gt;V600E/K&lt;/sup&gt;</strong></td>
<td>Dabrafenib (Tafinlar)</td>
<td>THxID-BRAF CDx Test&lt;sup,#,NHS&lt;/sup&gt; (BioMérieux Inc)</td>
<td>Indicates the presence of V600E and V600K mutations in patients with unresectable or metastatic melanoma to select those who may benefit from treatment with dabrafenib and trametinib (or cobimetinib)</td>
</tr>
<tr>
<td><strong>MEK</strong></td>
<td>Trametinib (Mekinist)</td>
<td>Cobimetinib*** (Cotellic)</td>
<td></td>
</tr>
<tr>
<td><strong>PD-L1</strong></td>
<td>Pembrolizumab (Keytruda)</td>
<td>See entry for pembrolizumab in Table 2.</td>
<td>See entry for pembrolizumab in Table 2</td>
</tr>
<tr>
<td></td>
<td>Nivolumab (Opdvo)</td>
<td>See entry for nivolumab in Table 2.</td>
<td>See entry for nivolumab in Table 2</td>
</tr>
</tbody>
</table>
*** agents that have been removed from National Institute for Health and Care Excellence (NICE) guidance or rejected by NICE

NHS companion diagnostic tests which are used in the NHS based on cost/benefit ratio

nNHS companion diagnostic tests which are not used in the NHS based on cost/benefit ratio

In 2013 the FDA approved the use of dabrafenib and trametinib in combination alongside the THxID BRAF test from BioMérieux. It is the second companion diagnostic approved by the FDA for BRAF mutation detection following approval of Roche’s Cobas 4800 BRAF V600 Mutation Test in 2011

Table 5: Biomarkers important in ovarian cancer, the precision medicine (PM) agents used in this disease and the companion diagnostic (CDx) tests used to select patients most likely to respond

<table>
<thead>
<tr>
<th>Biomarkers</th>
<th>Agents used</th>
<th>Examples of companion diagnostic tests</th>
<th>Use of test results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mutated BRCA1 and BRCA2</td>
<td>Niraparib (Zejula)</td>
<td>BRACAnalysis CDx NHS (Myriad Genetics Inc)</td>
<td>Identifies ovarian cancer patients carrying germline BRCA mutations who should respond to niraparib</td>
</tr>
<tr>
<td>Olaparib (Lynparza)</td>
<td>BRCA Analysis CDx NHS (Myriad Genetics Inc)</td>
<td>Identifies ovarian cancer patients carrying germline BRCA mutations who may benefit from treatment with olaparib</td>
<td></td>
</tr>
<tr>
<td>Rucaparib (Rubraca)</td>
<td>FoundationFocus CDxBRAC NHS (Foundation Medicine Inc)</td>
<td>Identifies advanced ovarian cancer patients with mutations in their BRCA1 and BRCA2 genes who are likely to benefit from treatment with rucaparib</td>
<td></td>
</tr>
<tr>
<td>VEGFR1 and VEGFR2</td>
<td>Bevacizumab (Avastin)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td>CA125 and CA125 II</td>
<td>None</td>
<td>Test kits, based on ELISA, are under development in collaboration between Morphotek Inc and Fujirebio Diagnostics</td>
<td>CA125 and CA125 II are diagnostic biomarkers for ovarian cancer</td>
</tr>
</tbody>
</table>

NHS companion diagnostic tests which are used in the NHS based on cost/benefit ratio

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Table 6: Biomarkers important in prostate cancer, the precision medicine (PM) agents used in this disease and the companion diagnostic (CDx) tests used to select patients most likely to respond

<table>
<thead>
<tr>
<th>Biomarkers</th>
<th>Agents used</th>
<th>Examples of companion diagnostic tests</th>
<th>Use of test results</th>
</tr>
</thead>
<tbody>
<tr>
<td>AR (Androgen Receptor)</td>
<td>Apalutamide* (Erleada)</td>
<td>Numerous commercial test kits available.</td>
<td>There is currently no screening program for prostate cancer in the UK, as benefits have not been proven to outweigh the risks.</td>
</tr>
<tr>
<td></td>
<td>Abiraterone (Zytiga)</td>
<td>-</td>
<td>N/A</td>
</tr>
<tr>
<td></td>
<td>Enzalutamide (Xtandi)</td>
<td>-</td>
<td>N/A</td>
</tr>
<tr>
<td></td>
<td>Darolutamide** (Nubeqa)</td>
<td>-</td>
<td>N/A</td>
</tr>
<tr>
<td>Mutated BRCA1 and BRCA2</td>
<td>Olaparib** (Lynparza)</td>
<td>See Table 5 for examples of companion diagnostic tests available for BRCA1 and BRCA2</td>
<td>BRACA1/2 analysis has been used experimentally to identify prostate cancer patients who are likely to respond to PARP inhibitors such as olaparib</td>
</tr>
</tbody>
</table>

* agents that are likely to be approved by National Institute for Health and Care Excellence (NICE) in the near future, but may not necessarily be used in the NHS

** agents that are unlicensed in the UK at the time of writing

Table 7: Biomarkers important in renal cancer, the precision medicine (PM) agents used in this disease and the companion diagnostic (CDx) tests used to select patients most likely to respond

<table>
<thead>
<tr>
<th>Biomarkers</th>
<th>Agents used</th>
<th>Examples of companion diagnostic tests</th>
<th>Use of test results</th>
</tr>
</thead>
<tbody>
<tr>
<td>VEGFR 1–3 and PDGFRα/β</td>
<td>Pazopanib (Votrient)</td>
<td>NexCourse*NHS Complete/Solid Genoptix-A (Novartis Inc)</td>
<td>Over-expression of VEGFR 1–3 is detected and allows the selection of patients for treatment with Pazopanib</td>
</tr>
<tr>
<td></td>
<td>Sunitinib (Sutent)</td>
<td>LC-MS/MS*NHS</td>
<td>Within the NHS, plasma levels of the active metabolite of Sunitinib (i.e. N-</td>
</tr>
</tbody>
</table>
desmethylsunitin-ib) are measured to assess adherence and optimise treatment

<table>
<thead>
<tr>
<th>Biomarkers</th>
<th>Agents used</th>
<th>Examples of companion diagnostic tests</th>
<th>Use of test results</th>
</tr>
</thead>
<tbody>
<tr>
<td>VEGFR 1-3</td>
<td>Axitinib (Inlyta)</td>
<td>Sorafenib by LC-MS/MS\textsuperscript{NHS}</td>
<td>Measures the active metabolite of Sorafenib (Sorafenib N-oxide) in plasma to assess adherence, monitor toxicity and optimise treatment</td>
</tr>
<tr>
<td></td>
<td>Cabozantinib (Cabometyx)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td>PD-L1</td>
<td>Pembrolizumab* (Keytruda)</td>
<td>See entry for pembrolizumab in Table 2</td>
<td>See entry for pembrolizumab in Table 2</td>
</tr>
<tr>
<td></td>
<td>Nivolumab (Opdivo)</td>
<td>See entry for nivolumab in Table 2</td>
<td>See entry for nivolumab in Table 2</td>
</tr>
<tr>
<td></td>
<td>Atezolizumab** (Tecentriq)</td>
<td>None</td>
<td>-</td>
</tr>
</tbody>
</table>

* agents that are likely to be approved by National Institute for Health and Care Excellence (NICE) in the near future, but may not necessarily be used in the NHS\textsuperscript{40,71}  
** agents that are unlicensed in the UK at the time of writing  
\textsuperscript{NHS} companion diagnostic tests which are used in the NHS based on cost/benefit ratio\textsuperscript{72,73}  
\textsuperscript{nNHS} companion diagnostic tests which are not used in the NHS based on cost/benefit ratio\textsuperscript{72,73}  

Table 8: Biomarkers important in hepatocellular carcinoma, the precision medicine (PM) agents used in this disease, and the companion diagnostic (CDx) tests used to select patients most likely to respond
Table 9: Biomarkers used in medullary and papillary thyroid cancers, the precision medicine (PM) agents used in this disease, and the companion diagnostic (CDx) tests used to select patients most likely to respond

<table>
<thead>
<tr>
<th>Biomarkers</th>
<th>Agents used</th>
<th>Examples of companion diagnostic tests</th>
<th>Use of test results</th>
</tr>
</thead>
<tbody>
<tr>
<td>VEGFR and PDGFR</td>
<td>Sorafenib (Nexavar)</td>
<td>See entry for Sorafenib in Table 8</td>
<td>See entry for Sorafenib in Table 8</td>
</tr>
<tr>
<td></td>
<td>Lenvatinib (Lenvima)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Regorafenib** (Stivarga)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td>VEGFR</td>
<td>Cabozantinib (Cometriq)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Vandetanib** (Caprelsa)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td>PD-L1</td>
<td>Nivolumab** (Opdivo)</td>
<td>See entry for nivolumab in Table 2</td>
<td>See entry for nivolumab in Table 2</td>
</tr>
</tbody>
</table>

** agents that are unlicensed in the UK at the time of writing

NHS companion diagnostic tests which are used in the NHS based on cost/benefit ratio

** agents that are unlicensed in the UK at the time of writing
Table 10: Biomarkers used in pancreatic neuroendocrine tumours (PNTs), the precision medicine (PM) agents used in this disease, and the companion diagnostic (CDx) tests used to select patients most likely to respond

<table>
<thead>
<tr>
<th>Biomarkers</th>
<th>Agents used</th>
<th>Examples of companion diagnostic tests</th>
<th>Use of test results</th>
</tr>
</thead>
<tbody>
<tr>
<td>VEGF</td>
<td>Everolimus (Afinitor)</td>
<td>Tandem mass spectrometric (LC-MS/MS) assay for measuring everolimus&lt;sup&gt;NHS&lt;/sup&gt;</td>
<td>Due to a narrow therapeutic index, plasma concentrations of everolimus must be routinely monitored, usually on a pre-dose (trough) sample</td>
</tr>
<tr>
<td>VEGFR, VEGFR2 and PDGFRα/β</td>
<td>Sunitinib (Sutent)</td>
<td>See entry for Sunitinib in Table 7</td>
<td>See entry for Sunitinib in Table 7</td>
</tr>
<tr>
<td></td>
<td>Vandetanib** (Caprelsa)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Erlotinib** (Tarceva)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td>UGT1A1</td>
<td>Nano-Liposomal Irinotecan*** (Onivyde)</td>
<td>See entry for Irinotecan in Table 3</td>
<td>See entry for Irinotecan in Table 3</td>
</tr>
<tr>
<td>BRCA1 and BRCA2</td>
<td>Olaparib* (Lynparza)</td>
<td>See Table 5 for examples of companion diagnostic tests available for BRCA1 and BRCA2</td>
<td>See Table 5 for examples of companion diagnostic tests available for BRCA1 and BRCA2</td>
</tr>
</tbody>
</table>

** agents that are unlicensed in the UK at the time of writing
*** agents that have been removed from National Institute for Health and Care Excellence (NICE) guidance or rejected by NICE

<sup>NHS</sup> companion diagnostic tests which are used in the NHS based on cost/benefit ratio<sup>72,73</sup>

Table 11: The main biomarker used in bladder cancer, the precision medicine (PM) agents used in this disease, and the companion diagnostic (CDx) tests used to select patients most likely to respond

<table>
<thead>
<tr>
<th>Biomarkers</th>
<th>Agents used</th>
<th>Examples of companion diagnostic tests</th>
<th>Use of test results</th>
</tr>
</thead>
<tbody>
<tr>
<td>PD-L1</td>
<td>Durvalumab** (Imfinzi)</td>
<td>See entry for Durvalumab in Table 2</td>
<td>See entry for Durvalumab in Table 2</td>
</tr>
<tr>
<td></td>
<td>Atezolizumab** (Tecentriq)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td>Biomarker</td>
<td>Agents used</td>
<td>Examples of companion diagnostic tests</td>
<td>Use of test results</td>
</tr>
<tr>
<td>---------------------------</td>
<td>----------------------------------</td>
<td>----------------------------------------</td>
<td>---------------------</td>
</tr>
<tr>
<td>Hedgehog signaling pathway</td>
<td>Vismodegib (Erivedge)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Sonidegib** (Odomzo)</td>
<td>None</td>
<td>-</td>
</tr>
</tbody>
</table>

** agents that are unlicensed in the UK at the time of writing

Table 12: The main biomarker relevant to basal cell carcinoma, and the small molecule inhibitors associated with it

** agents that are unlicensed in the UK at the time of writing

<table>
<thead>
<tr>
<th>Biomarkers</th>
<th>Agents used</th>
<th>Examples of companion diagnostic tests</th>
<th>Use of test results</th>
</tr>
</thead>
<tbody>
<tr>
<td>C-Kit (also known as CD117) or DOG1</td>
<td>Imatinib (Glivec)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Sunitinib (Sutent)</td>
<td>See entry for Sunitinib in Table 7</td>
<td>See entry for Sunitinib in Table 7</td>
</tr>
<tr>
<td></td>
<td>Regorafenib (Stivarga)</td>
<td>None</td>
<td>-</td>
</tr>
<tr>
<td>PDGFRA</td>
<td>Sunitinib (Sutent)</td>
<td>See entry for Sunitinib in Table 7</td>
<td>See entry for Sunitinib in Table 7</td>
</tr>
<tr>
<td></td>
<td>Regorafenib (Stivarga)</td>
<td>None</td>
<td>-</td>
</tr>
</tbody>
</table>
Table references


