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High-throughput molecular assays for inclusion in personalised oncology trials – State-of-the-art and beyond

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In the last decades, the development of high-throughput molecular assays has revolutionised cancer diagnostics, paving the way for the concept of personalised cancer medicine. This progress has been driven by the introduction of such technologies through biomarker-driven oncology trials. In this review, strengths and limitations of various state-of-the-art sequencing technologies, including gene panel sequencing (DNA and RNA), whole-exome/whole-genome sequencing and whole-transcriptome sequencing, are explored, focusing on their ability to identify clinically relevant biomarkers with diagnostic, prognostic and/or predictive impact. This includes the need to assess complex biomarkers, for example microsatellite instability, tumour mutation burden and homologous recombination deficiency, to identify patients suitable for specific therapies, including immunotherapy. Furthermore, the crucial role of biomarker analysis and multidisciplinary molecular tumour boards in selecting patients for trial inclusion is discussed in relation to various trial concepts, including drug repurposing. Recognising that today's exploratory techniques will evolve into tomorrow's routine diagnostics and clinical study inclusion assays, the importance of emerging technologies for multimodal diagnostics, such as proteomics and in vivo drug sensitivity testing, is also discussed. In addition, key regulatory aspects and the importance of patient engagement in all phases of a clinical trial are described. Finally, we propose a set of recommendations for consideration when planning a new precision cancer medicine trial.

Keywords: biomarkers, clinical trials, omics technologies, personalised oncology, precision cancer medicine, precision diagnostics

Introduction

Personalised medicine, also referred to as precision medicine, has become increasingly important in cancer care. The concept, defined by the European Commission as 'using phenotypes and genotypes for tailoring the right therapeutic strategy for the right person at the right time' [1], is intimately linked to the plethora of novel options for the treatment of cancer, many of them targeted to specific molecular alterations [2]. To predict responses to these targeted treatments, a range of diagnostic tools are needed, most of them being molecular tests [3].

The first wave of treatment-predictive assays included in situ hybridisation techniques that detected gene amplification or rearrangements or surrogate immunohistochemistry visualising protein overexpression. *ERBB2* (*HER2/neu*) amplification, originally a target for antibody treatment in breast cancer, is the prime example in this context [4–6]. The next wave of assays used targeted analyses in the form of, for example real-time quantitative polymerase chain reaction (PCR), allele-specific PCR or droplet digital PCR (ddPCR) [7–10]. These are reliable, sensitive assays, widely used for single gene testing but with limited capacity for multiplexing and will not be described further.

The first generation of sequencing, the so-called Sanger sequencing [11], was for a long time the gold standard. However, given the limitations in throughput, in detecting low allele frequencies and in handling low-quality nucleic acids, it has largely been replaced by high-throughput techniques based on massive parallel sequencing, often termed next-generation sequencing (NGS) that allows interrogation of many or all, genes at the same time [12].

In parallel, there has been a rapid development of testing strategies for trial inclusion and personalised oncology trial designs [13]. After a long period of clinical trials based on single-gene testing, multigene sequencing is now the norm. An important family of trials, widening the scope of molecular profiling even further, is based on the repurposing of targeted treatments in cancer types outside the current indication (TAPUR [14], DRUP [15]). To include patients in these studies, assessing the full range of clinically actionable molecular alterations known to date is needed.

As part of the ongoing Personalised Cancer Medicine for all EU citizens (PCM4EU) project [16], we reviewed diagnostic tools currently used for including patients in personalised oncology trials and compared their strengths and weaknesses. Key aspects affecting assay choice, as well as the rapidly evolving fields of complex biomarkers and liquid biopsies, are also highlighted. In addition, the assays are set in the context of the multidisciplinary decision-making for study inclusion at molecular tumour boards (MTBs), using clinical decision support systems (CDSSs) to integrate different resources and interpret the clinical relevance of the sequencing results [17]. Finally, current trends in assay development and emerging technologies are discussed to assist in the long-term planning of the diagnostic set-up used for enrolment in personalised oncology trials.

Gene panel sequencing – the new gold standard in cancer genomics

Gene panels are based on fixed assay designs assessing a specific set of genes or other biomarkers and can be divided by the chemistry used to target specific gene regions and by panel size. Amplicon-based panels have been widely used in cancer diagnostics, capitalising on their ability to assess biomarkers from DNA or RNA of lower quantity, which is common in formalin-fixed paraffin-embedded (FFPE) diagnostic samples. The use of amplicons of limited stretches of nucleic acids is perfectly suited for variant calling in the hotspot regions that define many of the activating mutations used for treatment selection. However, the robustness comes at the cost of sequence information limited to the tested regions and amplification biases, and at the risk of interaction between the primers used in large designs. This limits the flexibility to adapt or expand the panel design.

In contrast, capture-based panels use sets of capture probes, also called baits, to pull down regions of interest. This enables sequencing longer stretches of DNA or RNA to better analyse GC-rich regions and provide more reliable calling of copy-number alterations (CNAs). Additionally, new probes can be added without compromising quality metrics as the probes do not interact in the same way as primers might do. On the other hand, a higher sample input is generally needed, and sequencing of low-quality samples may prove more challenging.

Driven by (i) the increase in a number of clinically relevant genes, (ii) the introduction of tumour-agnostic indications and (iii) the addition of complex biomarkers that necessitate the interrogation of larger genomic footprints, the scope has over time shifted towards pan-cancer designs and increasing panel sizes for clinical diagnostics and study inclusion. When gene panels were introduced, amplicon-based designs of 10–50 genes predictive of targeted treatments – sometimes in a cancer type-specific manner – dominated. This approach has been preferred in many molecular pathology laboratories due to the ability to analyse fewer samples and utilise bench-top sequencers at a low cost and within a short time frame (days). As a

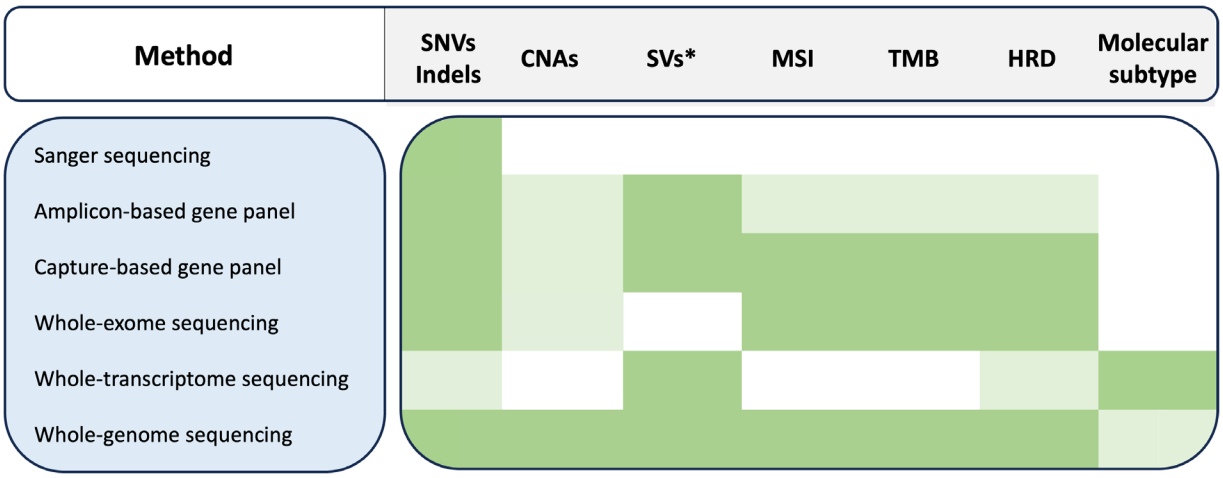

Fig. 1 *Detection capacity of different sequencing-based assays. SNVs, single nucleotide variants, Indels, insertion/deletions, CNAs, copy-number alterations, MSI, microsatellite instability, TMB, tumour mutational burden, HRD, homologous recombination deficiency. *Excludes CNAs.*

next step, both academic institutions [18] and diagnostic companies introduced substantially larger gene panels covering a range of 300–600 genes (equivalent to ~1 Mb). The goal was to achieve comprehensive genomic profiling independent of the cancer type, including all druggable gene variants and genes deemed cancer-relevant in a clinical context, while also allowing for exploratory biomarkers. Different gene panels are often utilised for solid tumours and haematological malignancies, as the spectrum of clinically relevant genetic aberrations differs. Another important aspect of the larger genomic coverage is the ability to interrogate complex biomarkers, such as microsatellite instability (MSI), tumour mutation burden (TMB) and homologous recombination deficiency (HRD) (Fig. 1).

An important variation of panel sequencing is targeted RNA sequencing, adding sensitivity and reducing cost in fusion gene detection compared to more traditional in situ or cytogenetic analyses. As breakpoints often are found in large and repetitive intronic regions, the ability to amplify or pull down the fusion gene transcripts reduces the number of sequenced bases and minimises mapping errors. The fact that these transcripts often are overexpressed also adds sensitivity to the fusion gene calling. Although amplicon-based approaches in targeted RNA sequencing can only detect predefined fusions, gene panels based on hybrid capture or single primer extension enable the detection of all selected fusion genes, given that baits against at least one of the fusion partners are included in the design [19, 20].

Moving towards genome-wide technologies

Whole-exome sequencing (WES) enables the interrogation of the estimated ~20,000 genes in the genome [21]. Similar to gene panel sequencing, this is a targeted approach whereby all coding genes are enriched and sequenced and usually performed with a paired germline sample (often peripheral blood). Initially, WES was mainly used for research purposes, characterising the genomic landscape of most cancer types [22]. Once WES was demonstrated to work on FFPE material [23], it became a feasible approach for clinical diagnostics. Using this approach, single nucleotide variants (SNVs) and small indels, as well as complex markers (MSI, TMB and HRD), can be readily identified using a sequencing depth of 100–300x (Fig. 1) [24, 25]. Even though copy-number variant-calling is better than in gene panels, WES is suboptimal as compared to *whole-genome sequencing* (WGS) and has a limited ability to detect other structural variants (SVs). To avoid the need for continuous updates of gene panels, WES is increasingly applied in personalised cancer trials and in routine practice at some cancer centres, particularly for paediatric cancer [26].

WGS is the most comprehensive genomic assay to detect all types of genetic alterations, including SNVs/indels, CNAs and SVs. In comparison

to WES, WGS can assess mutation signatures (MSI, TMB and HRD) with higher precision [27–29]. However, it is still difficult to perform WGS on FFPE material, which has limited its broad-scale clinical implementation. Of note, WGS requires substantial investments in large-scale sequencing and computational infrastructure, which also challenge broad adoption in healthcare [30]. Compared to WGS in rare diseases, for which a sequence depth of 30*x* is applied, tumour samples are sequenced to at least 90*x* to allow the detection of genetic alterations in at least 20% tumour cell purity. This limits the ability to detect subclonal variants, which can be determinants for therapy and/or trial opportunities. Given the large number of variants detected by WES and WGS, the sequencing of a matched normal sample is generally included to identify tumour-specific events. This also allows for the identification of pathogenic germline variants present in approximately 10% of cancer patients [31, 32] but contributes to the overall high sequencing cost, although the price for sequencing is steadily decreasing [27].

Nevertheless, for certain cancer types and defined clinical indications, such as haematological malignancies, sarcomas and rare cancers, WGS is under implementation in routine diagnostics, often through national initiatives, for example in England, France, Germany, Denmark, Norway and Sweden [33–36]. In paediatric cancer, WGS has diagnostic, risk-stratifying and predictive impact, as well as the ability to detect germline predisposition. In a recent Swedish study, WGS provided clinically relevant information in >90% of patients with paediatric solid tumours while adding new information in half of patients and contributed to the revision of the diagnosis in a few cases [37]. In most studies including haematological malignancies, an almost perfect match with the findings of standard-of-care assays has been reported while also providing new clinically important information in a proportion of patients [38, 39]. In sarcomas, which consist of around 100 entities [40], WGS significantly improves the diagnostic procedure and identifies treatment targets, and, as evidenced by the Dutch experience, the WGS workflow can be integrated in routine diagnostics [41].

WGS is also included in personalised oncology trials, primarily as an explorative analysis, whereas some studies have also applied it for study inclusion. For instance, a prospective trial using WGS in rare cancers ($n = 1310$) provided treatment recommendations for >85% of patients [33], whereas, in another trial utilising WGS, one third of patients with metastatic disease demonstrated a clinical benefit [42].

Beyond response prediction, recent studies suggest that WGS will support the transition from organ-centric to molecular-based classifications of cancer and complement diagnostic strategies for, for example patients with cancers of unknown primary [29, 43, 44].

Whole-transcriptome sequencing (WTS) or RNA sequencing is used to detect gene fusions as well as to identify gene expression signatures linked to certain disease (sub)types, drug targets or clinical outcomes. Protocols have primarily been developed for RNA prepared from fresh-frozen samples, although protocols have been established for FFPE samples more recently [45]. WTS is often performed together with WES/WGS, for example in haematological malignancies and sarcomas, to confidently identify fusion gene transcripts and aid disease subclassification [46]. An increase in usage is predicted, in particular in its capacity to detect gene expression profiles linked to treatment (e.g. poly(ADP)-ribose polymerase (PARP) inhibitors).

Refining diagnostics using array-based technologies

High-resolution genomic arrays, that is single nucleotide polymorphism- or oligo-arrays, are applied to detect cancer-specific CNAs, that is deletions and amplifications, whereas they generally cannot provide information on other SVs (Fig. 1). Genomic arrays have been specifically developed to allow the study of FFPE specimens, though the background noise level is slightly higher than sequence-based approaches. Genomic arrays are still applied in diagnostics of certain malignancies, particularly for paediatric leukaemia and central nervous system (CNS) tumours, although it is foreseen that they will be replaced by WGS/WES once they are implemented in standard-of-care.

Using gene expression arrays, different gene signatures have been identified within certain tumour types, some of them linked to clinical outcomes or specific treatment selection. For instance, in breast cancer and lymphoma, several subtypes were defined early on and significantly improved patient risk stratification [47, 48]. In fact, different types of gene expression-based assays utilising different technologies have been developed

for breast cancer (e.g. Prosigna (NanoString), Oncotype Dx (quantitative-PCR) and MammaPrint (array)) to predict patient response to treatment and recurrence-free survival. Such assays have also been used for patient selection and/or stratification for clinical trials in breast cancer [48, 49].

In the last 10–15 years, it became evident that tumours demonstrate characteristic DNA methylation profiles, often linked to the cell-of-origin, that could be used for the subclassification of certain tumour entities. As more comprehensive DNA methylation arrays were developed (such as the EPIC array), they were applied as routine methods to classify histologically heterogeneous tumour types, such as paediatric patients with CNS tumours [50, 51]. In this way, methylation arrays have contributed significantly to the diagnostic procedure by reducing the risk of making an incorrect diagnosis. Today, DNA methylation arrays are considered standard-of-care for cancer-type prediction in this patient group. The recent improvements in long-read sequencing technologies now open up for assessment of DNA methylation profiling by sequencing.

Gene signatures as biomarkers in clinical trials

Cancer genomes are influenced by endogenous (e.g. defective DNA repair) and/or exogenous (e.g. cigarette smoking and UV-light) mutational processes. This creates different mutational patterns, often referred to as 'genetic scars', and so far, 65 different signatures have been linked to specific biological processes, such as BRCA-deficiency (SBS3), mismatch repair deficiency (MMRd, SBS26 and SBS44), age at diagnosis (SBS1) or smoking (SBS4) and UV-light (SBS7) exposure [52, 53]. Mutational signatures are an important addition to the biomarker repertoire, but their use faces several challenges [53]. First, the identification of a signature depends on the methodology and the algorithms used for data analysis. Second, most of them represent a score on a continuous scale, and definitions of 'negative' and 'positive' samples are not straightforward. Third, as the underlying cause for each of them can be distortions of one of several genes/proteins, matching appropriate therapeutic intervention is not necessarily intuitive.

Today, the most established signatures in the clinical setting are MMRd, HRD and the TMB. MMRd tumours display dysfunctional repair of replication errors such as base–base and indel mismatches. Traditionally, this is identified by targeted molecular analyses evaluating MSI and/or lack of expression of MMR proteins by immunohistochemistry. Substituting these techniques with larger gene panels and WES/WGS has not been trivial and requires validation [54]. For instance, probes have been added to a capture-based panel to identify MSI in a tumour by applying algorithms suitable for NGS-derived data (e.g. MSIsensor, MSIseq). At present, MMRd/MSI measurements are obligate in most personalised oncology trials as a biomarker for immune checkpoint inhibitors.

The level of TMB is known to vary between tumour entities [55], and different strategies have been applied to measure TMB using gene panels, WGS and WES. Most MMRd tumours show a high number of mutations, but a high TMB can be caused by other mechanisms, including UV exposure, cigarette smoking or mutations in other components of the DNA repair machinery. TMB is defined by the total number of somatic variants in a defined region and can be calculated in various ways. The field is still in a phase of algorithm development, and cut-off values need to be validated [56].

For HRD analysis, which relates to deficiency in repairing double-strand breaks through homologous recombination, a few commercial and clinically validated assays are available. Patients with HRD-positive tumours, predominantly ovarian, breast, prostate and pancreatic cancers, have a higher probability of responding to PARP inhibitors and platinum-containing regimens. Today, particularly one test (myChoice CDx from Myriad Genetics) has been used in clinical trials, leading to FDA approval [57], but large harmonisation efforts are ongoing to secure adequate HRD measurements [58], and other complex markers are foreseen to be developed [59, 60]. Algorithms have been developed to assess HRD from WGS/WES data, and efforts are made to accommodate this measurement in larger gene panels.

Overall, it is paramount to include these complex markers in the testing armoury, especially as there is a need for evidence concerning the tumour type-specific role for each of them [61]. However, it is important to keep in mind that there is still a lack of harmonisation of the different classifiers used for study inclusion based on these measurements.

Liquid biopsies for biomarker detection

Multiple challenges are faced by comprehensive genomic profiling of tumour tissue: (i) High-quality tissue biopsies are challenging to secure; (ii) some tumours/metastases are not accessible; (iii) tumour/metastases can be heterogeneous and molecular changes in one biopsy/location may not represent the biology of the disease; and (iv) multiple biopsies during the treatment course are difficult to secure due to the invasiveness of the procedure [62]. As a result, the use of liquid biopsies has attracted increasing interest. Liquid biopsies are minimally invasive samples from body fluids that can be used for the detection of cancer-derived molecules (i.e. DNA, RNA or proteins). Most explored is peripheral blood, where the presence and level of cell-free circulating tumour DNA (ctDNA) in plasma are measured using various methodologies (NGS-based or ddPCR).

Detection of ctDNA in plasma can improve early cancer detection, therapy guidance and longitudinal disease monitoring. Still, the implementation is not without challenges. As non-malignant cells also release cell-free circulating DNA (cfDNA), the tumour fraction (ctDNA) is highly variable and often consists of <1% of the cfDNA [63]. Further, due to its mainly apoptotic origin, cfDNA is highly fragmented, which complicates downstream analyses [64]. Finally, to detect ctDNA, tumour-specific alterations must be identified by a sensitive assay. As the DNA mutation spectra of cancer are highly variable across tumours and some tumours may bear infrequent genomic alterations, targeted sequencing of selected loci may not capture all variants present in a given case.

Liquid biopsy analysis can be applied to identify SNVs/indels and gene fusions, whereas CNAs and complex markers (HRD, MSI and TMB) can be more challenging. Targeted analysis using pre-defined targeted sequencing or ddPCR is most common, with the latter being more sensitive but including fewer targets. There is generally a high concordance in molecular results obtained using tissue and plasma samples, although this may vary depending on tumour type and disease stage. Furthermore, pre-analytical factors, such as transport and storage conditions, stabilisation reagents and extraction protocols, significantly affect the integrity and yield of cfDNA [65, 66]. The need for high sensitivity to detect ctDNA using NGS-based techniques also requires deep sequencing, resulting in a high cost per sample. Moreover, the detection of variants related to clonal haematopoiesis of indeterminate potential, a phenomenon of the ageing immune system, may influence the interpretation of the variants identified [67].

The use of comprehensive molecular profiling for trial matching also allows for disease monitoring, measuring therapy response and measurable residual disease. For these applications, tumour-informed approaches (with analyses adapted to tumour type or personalised for the individual tumour) can be used to cost-effectively achieve high sensitivity [68].

To summarise, liquid biopsies are used more and more in clinical trials, mainly to increase the number of patients screened for study inclusion. For instance, in many DRUP-like trials, inclusion based on ctDNA analysis is allowed when a tumour biopsy is not available [69]. However, to widely adopt liquid biopsy in clinical practice, more interventional clinical trials, as well as standardisation of methods and interpretation, are necessary [70].

Considerations on selecting molecular assays for study inclusion

It is evident that high-throughput molecular assays have an increasingly important role in selecting patients for clinical trials. Nevertheless, as this is a rapidly evolving field, it is important to have up-to-date knowledge of the different techniques' strengths, weaknesses and applicability when setting up a personalised oncology trial.

In traditional randomised trials, the impact of a new drug or combination of drugs is evaluated by comparing the effect of the intervention versus the control arm. If a biomarker assessment is included to define the study population, a specific molecular assay or set of biomarkers is defined in the study protocol. However, in personalised oncology trials based on the basket and/or umbrella concept, a more comprehensive analysis is needed to allow the interrogation of a larger number of biomarkers, and here, broad gene panel sequencing is the preferred choice. This enables the detection of various types of genetic aberrations (SNVs/indels, CNAs and SVs), whereas the large gene content also enables the assessment of more complex biomarkers (HRD, TMB and MSI) (Fig. 1). Usually, panel sequencing is performed on a new tumour biopsy before study inclusion, although the assessment of

biopsies taken up to 6 months prior to inclusion is considered acceptable. As mentioned, the use of comprehensive genomic profiling by ctDNA analysis has, in recent years, also increased the number of screened patients, as it is not always possible to take a new tissue biopsy. However, although ctDNA analysis works well to detect predefined alterations in individual genes, the assessment of complex biomarkers, such as the HRD status, is still in its infancy.

Despite using panel sequencing to identify patients for study inclusion, many of the personalised oncology trials encompass more exploratory testing using genome-wide techniques such as WES, WGS and WTS. This enables in-depth investigations to find novel markers linked to therapy response or resistance. In more recent years, several studies, for example the MASTER trial in Germany, have started applying WGS to stratify patients [14, 15]. IMPRESS-Norway includes patients based on a large gene panel (TSO500) and performs WGS and WTS on patients included in treatment cohorts (~20% of tested patients) [69]. In this way, they can both assess actionable genetic aberrations and have the possibility to find additional alterations beyond the genes included in larger gene panels.

In Table 1, a more detailed list of pros and cons of the different types of molecular assays is provided that can assist when deciding the strategy for a planned personalised oncology study. First, it is important to know if the assay of choice is compatible with FFPE tissue and what type of aberrations each assay can (and cannot) detect. For instance, to reliably assess complex biomarkers based on gene signatures, a general rule is that the larger the gene content, the more reliable is the information. The selection between sequencing strategies is also highly dependent on the availability of infrastructure. Most university hospitals have the resources and infrastructure to perform large panel/genome-wide sequencing, whereas regional hospitals may only have access to benchtop instruments and smaller panels.

Another important aspect is the timing of the genomic profiling. Although this is generally performed after all conventional treatment options are exhausted, an earlier assessment or 'pre-screening' would identify patients suitable for a clinical trial at an earlier stage. In this way, the time required for trial inclusion could be reduced significantly to the benefit of the patient.

Deciding on gene panel content might be challenging, and the use of databases gathering knowledge on genomic variant effects might be of use. As evidenced by a structured analysis of a meta-knowledgebase set up by the Variant Interpretation for Cancer Consortium, incorporating information from six well-established databases [71], it is challenging to establish consensus on clinical interpretations on a variant level, especially for biomarkers with lower evidence level. However, even though continued integration of databases is called for, the interpretation of gene variants and complex biomarkers of established relevance for tumour biology overlap considerably. In Fig. 2, data from the well-structured OncoKB database [72, 73], summing up data on 840 genes and 7636 alterations, illustrate currently relevant genes and biomarkers. In detail, OncoKB reports a total of 58 genes whose alterations act as biomarkers of drug response according to the higher evidence for actionability (i.e. levels 1, 2 and R1), which thus could be used in drug repurposing trials. An additional 19 genes are not yet clinical indications, but given compelling clinical or biological evidence for a treatment-predictive role, they might be used in clinical trials. To get a more complete picture, genes of importance for tumour biology also need to be taken into account. To this end, analyses of data from the Cancer Genome Atlas (TCGA) and the International Cancer Genome Consortium efforts [74, 75] could be used in assessing panel designs. In two TCGA papers summing up canonical signalling pathways [76] and genes significantly mutated in cancer [76], a set of genes likely to be explored for anti-cancer drug development can be found.

Next-generation multimodal diagnostics for trial inclusion

On the one hand, there is a need for improved predictive accuracy on diagnostics assays for the selection of responders to existing treatment strategies. On the other hand, there is also a need to broaden the biomarker-guided selection for patients whose tumours feature phenocopying of targetable pathways as well as develop biomarkers for targeted combination treatments in cancer trials [77]. This will require connecting the above-described genomic-based tools to molecular, cellular and functional phenotype analysis in diagnostics. As mentioned above, the European Commission's stated goal of 'using phenotypes and genotypes for tailoring the right therapeutic strategy' thrives towards a more comprehensive

Table 1. *Pros and cons with different high-throughput molecular assays.*

	Assay	FFPE compatible	Pros	Cons
Targeted analysis	Single gene analysis[a]	Yes	Specific	Low-throughput
	Amplicon-based gene panels	Yes	Lower amount of input DNA/RNA needed Lower quality of input DNA/RNA possible	Limited gene content Amplification biases
	Capture-based gene panels	Yes	Even sequencing coverage Detects additional types of aberrations (CNAs, SVs) Large designs allow interrogation of complex biomarkers (HRD, TMB, MSI)	Lower sequencing depth for gene panels with high gene content Reduced CNV detection for FFPE material
	Whole-exome sequencing	Yes	Detects SNVs/indels and CNAs Higher sequencing depth than WGS Increase the genomic region used to calculate complex biomarkers	Cannot detect SVs Reduced CNV detection for FFPE material
Array-based analysis	Gene expression arrays	Yes	Allows disease (sub)classification Detects cell-of-origin	Cannot detect fusion genes or other information provided by WTS
	SNP-arrays	Yes	Detects CNV and LOH Allows estimation of tumour purity and ploidy	Cannot detect SVs Reduced CNV detection for FFPE material
	DNA methylation arrays	Yes	Allows disease (sub)classification Detects cell-of-origin Allows estimation of tumour purity and ploidy	Only pre-selected targets detected
Genome-wide analysis	Whole-transcriptome sequencing (WTS)	Yes?	Detects fusion genes Disease (sub)classification Detects cell-of-origin Allows estimation of tumour purity and infiltrating cell populations	Less well established for FFPE material
	Whole-genome sequencing (WGS)	No	Detects all types of genomic aberrations including complex biomarkers Allows estimation of tumour purity	Not suitable for FFPE material Requires higher computing capacity Lower sequence depth than WES

Abbreviations: CNAs, copy-number alterations; FFPE, formalin-fixed paraffin-embedded; HRD, homologous recombination deficiency; LOH, loss of heterozygosity; MSI, microsatellite instability; SNP, single nucleotide polymorphism; SVs, structural variants; TMB, tumour mutation burden; WES, whole-exome sequencing.

[a]Includes different technologies that detect single gene/hotspot alterations such as fragment analysis, Sanger sequencing and others.

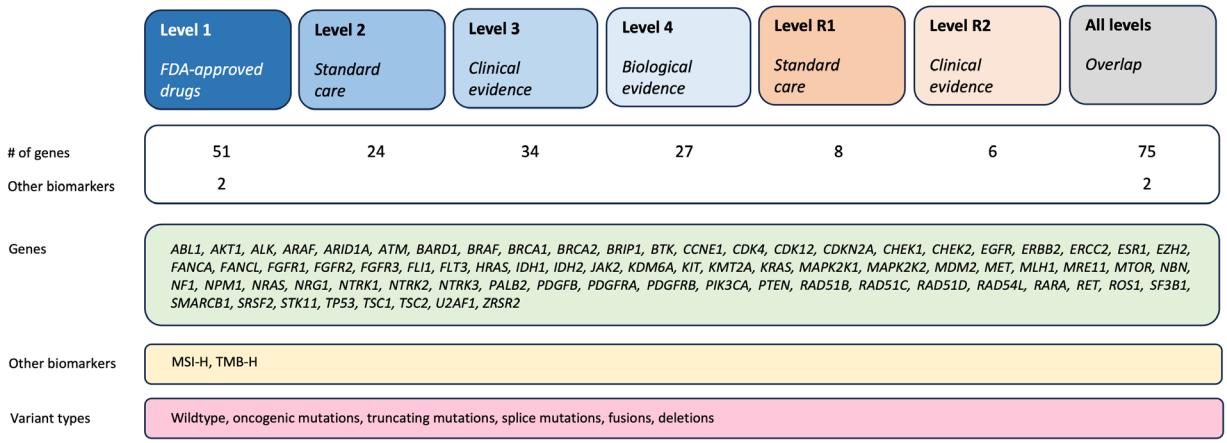

Fig. 2 *Levels of clinical evidence for genes and biomarkers categorised according to the OncoKB Therapeutic Level of Evidence v2.*

analysis of cancer drivers and immune evasion mechanisms by including molecular and imaging phenotype level information to support clinical decision-making. This can be done in a systematic and safe manner by integrating such biomarker developments as part of clinical trials, first by observational studies followed by actionable use of those biomarkers showing clinical validity in interventional trials.

As described, transcriptomics analysis has already been applied to profile tumour tissue for cancer subtyping; however, proteomics and metabolomics represent a more direct analysis of molecular phenotypes. Proteins compose virtually all drug targets used in personalised medicine. Hence, the analysis of the proteome has the potential to provide information on protein levels and their activation status by measuring the posttranslational modification status, provide information on protein networks in the context of an individual genome, as well as information on (sub)cellular and tissue location of drug targets and their interactomes. Protein level information has been readily used for diagnostics and treatment response prediction using individual protein biomarkers both in tissue samples, mainly by immunohistochemistry and in plasma samples using various assays. Despite this, comprehensive proteome analysis is still not used as a biomarker analysis for treatment selection in oncology. Moreover, the multimodal analysis of genome and proteome is largely underexplored for treatment selection as combination biomarkers. However, the rapid technological developments of mass spectrometry and affinity-based proteomics have opened new opportunities for proteogenomics in cancer diagnostics [78]. Today, comprehensive tumour cell proteomes can be analysed rapidly by mass spectrometry, providing a detailed molecular phenotypic view of each case [79–81]. Moreover, the combined use of genomics and proteomics data allows the detection of variant proteins and the impact of cancer-associated mutations on the protein level [82]. Today, cancer proteomics is widely used to study retrospective cohorts, whereas proteomics-driven biomarker-based clinical trials are lagging behind. To allow wider use of proteomics in clinical trials, the standardisation of sampling, quality procedures for robust analytical performance as well as clinical cut-offs for biomarkers and their reporting in a given trial need to be further developed.

Similarly, plasma proteome and metabolome analyses have great potential to predict treatment response and for early response evaluation. Both mass spectrometry and affinity-based proteomics methods can provide relevant data, not only from plasma but also from circulating blood components such as microvesicles and cells. Affinity-based methods like the proximity extension assay using antibodies and aptamer-based binding assay achieve very high-throughput plasma analysis, enabling large cohort-based studies [83]. However, adapting these affinity proteomics-based methods for clinical use and patient selection in a trial setting requires rigorous development to ensure that the right proteins are analysed regardless of individual background proteome changes.

Immunotherapy has profoundly changed the cancer treatment landscape and consequently, predictive biomarkers for patient selection are a hugely important area for future development. Combining genotype and phenotype level analysis is crucial as treatments such as immune checkpoint inhibitors modulate host–tumour interactions. Analysing intrinsic tumour characteristics, such as in MSI and TMB analyses, can only be considered surrogate markers for potential immune evasion mechanisms in connection to current treatments. Proteomics and metabolomics can instead provide direct analysis of important immune mediators in relation to clinical responses, whereas methods such as multiplex tissue marker analysis and spatial omics can add information about both immune cell infiltration and their molecular features [84]. In this area, image analysis by artificial intelligence (AI)-based methods has the potential to contribute significantly to future multimodal diagnostics as data volumes increase and methods develop. In general, AI tools are especially valuable in clinical trial contexts, from patient recruitment to biomarker analysis, but the field is in its infancy [85].

An interesting new data modality is the analysis of tumour-associated and tumour-specific antigens, also called neoantigens or neoepitopes. Here, integrated genomics, transcriptomics and proteomics analysis can provide valuable biomarker information [86], as well as open avenues for the therapeutic development of new modalities such as personalised cancer vaccines [87]. Radiomics is another rapidly developing biomarker modality considered in clinical trials [88]. In radiomics, quantitative image features are extracted from the data and can be developed to new biomarkers associated with outcome. Moreover, in this field, AI methods are entering the field [89].

Finally, functional phenotype analysis using ex-vivo drug screening has entered clinical trials as a selection tool in a few cutting-edge centres [90]. Here, patient-derived cells are tested for sensitivity to available cancer drugs for treatment selection in late-stage patients. Recently, the evaluation of drug combinations using ex-vivo screening has been incorporated in clinical trials (ComboMATCH (NCT05564377), EVIDENT (NCT0572520)). This approach also offers possibilities to gain knowledge on responders versus non-responders when carried out in parallel with other biomarker analysis. However, the requirements of clinical material, sophisticated workflows and cost are hindering the scalability of this approach beyond advanced research-intensive hospital settings.

In summary, multimodal diagnostics offers huge potential for personalised cancer medicine; however, new roadmaps for multimodal diagnostics development in connection to clinical trials are needed.

The importance of CDSS for patient inclusion

In the oncology setting, somatic variants must be evaluated for their pathogenicity and clinical actionability [91, 92]. A CDSS assists in this process by automating the annotation, classification and reporting of the observed variants, a complex process that is time-consuming and error-prone when performed manually. Examples of commercial CDSSs include Clinical Insight Interpret, IBM Watson for Genomics and OncoKDM, whereas academic CDSSs include Personal Cancer Genome Reporter [93] and the Molecular Tumour Board Portal [94]. As a core component of these CDSSs are databases that gather knowledge on the pathogenicity, diagnostic, prognostic and predictive relevance of genetic variants, such as ClinVar [95], OncoKB [72], CIViC [96] and The Clinical Knowledgebase [97]. A CDSS can also integrate other bioinformatic tools to estimate the relevance of variants whose effect has not yet been characterised [98]. Of note, an important output of the clinical actionability analysis is to identify matched drugs that are under evaluation in ongoing trials. However, available clinical trial databases, such as Clinicaltrial.gov (US-based) or EudraCT (European-based), do not follow a standardised model to collect the information on the molecular marker(s) used as inclusion (or exclusion) criteria. As a result, this information appears expressed in different manners and may be inexact, which prevents the CDSS from querying these resources with the accuracy required in clinical reporting. Therefore, local solutions have been developed to better address this process, like the Molecular Tumour Board Portal, which uses an in-house tool to match patient tumours and clinical characteristics with ongoing trial opportunities (Fig. 3). This technology, developed within Cancer Core Europe [99], is focused on the trials co-developed by the comprehensive cancer centres connected in the network; however, additional efforts are currently being made to extend the solution to a more comprehensive set of trials in Europe [100].

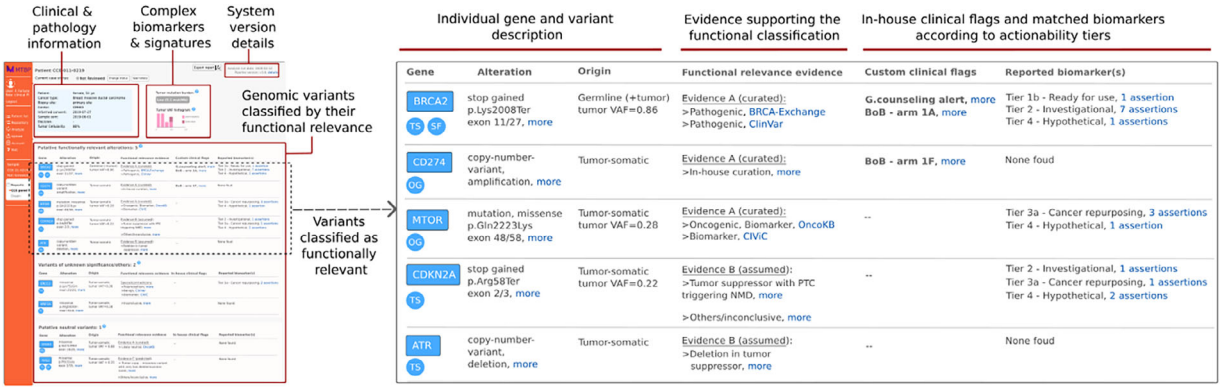

Fig. 3 *Example of a Molecular Tumour Board Portal (MTBP) report. The MTBP automates the interpretation and reporting of genomic variants in cancer, which requires assessing their functional relevance as well as their value as biomarkers of cancer diagnosis, prognosis and therapy response according to distinct levels of clinical evidence. The system also flags other events of interest, such as matched clinical trial opportunities and the presence of variants requiring genetic counselling. To do so, the MTBP integrates a number of knowledgebases and bioinformatic tools available in the community or developed in-house, as exemplified here, following a predefined – and thus consistent – process based on expert consensus. Of note, the reports generated by the system are interactive HyperText Markup Language (HTML) documents with comprehensive annotation aimed for expert review, such as that of a molecular tumour board. The results of this review will produce the final patient report, which contains the main conclusions of the clinical interpretation and treatment recommendation(s).*

With the increasing complexity of patient clinical and genomic data, and the availability of a larger number of relevant knowledgebases, AI will increasingly be applied to assist in guiding clinical choices in an MTB setting [101]. Several limitations exist, including quality and update frequencies of underlying data, variability in different populations, availability of medical records and extent and type of genomic data and drug availability, which are all aspects in need of improvement. So far, limited studies have been published on the relative performance of AI-based CDSS tools. A relevant metric to evaluate is the concordance between the recommendations provided by the AI-based method and the MTB, which varies from <60% to >90% as well as varies between tumour types [101]. Overall, although significant challenges still exist, CDSSs are key for an efficient implementation of precision oncology strategies, which greatly facilitate, but not substitute, the process of clinical interpretation provided by expert medical teams, such as that from an MTB.

Molecular tumour boards

Evaluation of eligibility and treatment decisions in personalised oncology trials depends on the complex integration of diagnostic and clinical information with results from comprehensive molecular profiling. Learnings from traditional multidisciplinary meetings have been a basis for the design of most MTBs. The MTB meeting most often comprises experts in oncology, pathology, radiology, molecular biology, bioinformatics and clinical genetics aiming at recommending the best treatment or trial option for a patient. According to van der Velden et al., some basic requirements are needed for an effective and operational MTB meeting: (i) harmonisation in cancer sequencing practices and procedures, (ii) minimal member and operational requirements and (iii) an appropriate unsolicited findings policy [15]. Most MTBs have experienced the need to have a defined structure while at the same time providing flexibility to facilitate the rapid change in biomarker testing, treatment options and trial opportunities [102]. The recent advances in integrative CDSS tools and virtual meeting solutions also influence the structure of MTBs [98, 103]. Finally, the design and content of a formal report of the molecular profiling results and MTB recommendations are not trivial, even in major comprehensive cancer centres, as a substantial number of physicians have low confidence in their ability to understand those reports [104]. Nevertheless, national/regional MTBs can also be a tool to strengthen equal access to testing and reduce inequity in personalised cancer diagnostics [105].

Regulatory aspects of molecular assays

Molecular testing for personalised oncology trial inclusion is guided by the same regulatory framework as clinical testing. An accreditation, typically according to the ISO 15189:2022 standard, guides the work to meet the necessary formal requirements. Quality assurance, both internal and external, for example by participation in external quality assessment schemes is vital for ensuring reliable results. One important change is the Regulation (EU) 2017/746 on in vitro diagnostic medical devices (IVDR), passed by the EU in April 2017 [106, 107]. The IVDR, which aims at patient safety, came into force on 26 May 2017. After a transition period of 5 years, implementation started in 2022 and occurs in several steps. The IVDR not only regulates the manufacture and placement on the market of industrially manufactured IVDs but also imposes conditions on the manufacture and use of in-house (IH)-IVDs for internal use by healthcare. From May 2022 onwards, the following conditions must be met

1. IH-IVDs must comply with the general safety and performance requirements (IVDR, Annex I).
2. The manufacture and use must take place within the EU.
3. IH-IVDs may only be used by the institution itself and may not be transferred to another legal entity.
4. Competent authorities shall be provided with relevant information on the devices upon request and shall have access to the health institutions to verify their activities.
5. IH-IVDs shall not be manufactured on an industrial scale.

Points 2–5 are easily addressed by most laboratories, whereas point 1 requires additional implementation. Article 5 (5) details the prerequisites and conditions for the use of IH-IVDs further: (i) The devices are not transferred to another legal entity, (ii) manufacture and use of the devices occur under appropriate quality management systems (due in 2024), (iii) the laboratory of the health institution is compliant with the ISO 15189 standard or where applicable national provisions (due in 2024) and (iv) the health institution justifies in its documentation that the target patient group's specific needs cannot be met by an equivalent device available on the market (due in 2028, so-called industry privilege). Importantly, the terms 'patient group's specific needs' and 'equivalent' are not definitely defined by the IVDR. The current prevailing legal view on these terms is an interpretation that considers the intention of the legislature, which integrates IH-IVDs as part of a spectrum of diagnostics (comprising both commercially available IVDR-labelled products and IH-IVDs) needed in daily clinical care. For example, patient-specific gene panels or assays to achieve conclusive test results when using low/impaired input material may qualify for the use of IH-IVDs.

In summary, the use of diagnostic tests requires compliance with the IVDR, but the IVDR explicitly allows for the use of IH-IVDs. Given the short innovation cycles in diagnostics and the need for rapid adaptation to new clinical settings, this approach is meaningful and justified. Appropriate and standardised validation and verification of IH-IVDs will play a crucial role in this context.

Public–private partnerships in investigator-initiated trials

Collaborative efforts between academia and industry facilitate the exchange of expertise, resources and data, expediting the development and testing of innovative therapies. In the realm of investigator-initiated trials, partnerships like these empower researchers to conduct trials that address specific clinical questions and explore unconventional therapeutic avenues, fostering a comprehensive approach to advancing oncology research and treatment. Such strong relations between academic institutions and industry are pivotal for advancing clinical trials in oncology, also in DRUP-like trials. Complementing trial approaches focusing on drug development, the aim of DRUP is to identify potential therapeutic benefits beyond a drug's approved purpose. This strategy can expedite the drug development process and offer new treatment options for patients with limited alternatives. The focus on repurposing existing drugs enhances efficiency and potentially reduces the time and costs associated with bringing new therapies to market.

Another multi-stakeholder example involving several industrial partners is Omico, a nationwide network of research and treatment centres in Australia. Omico plays a crucial role in promoting genomic cancer medicine by bringing together

major cancer centres, research institutes, government entities, industry partners and patients. Their focus on early detection and risk stratification contributes to personalised cancer risk management, ultimately improving the quality-of-life and survival rates for patients with advanced cancers. Cancer Core Europe, founded in 2014, is a legal alliance of seven comprehensive cancer centres formed to co-develop innovative cancer research and shared infrastructures. One of the pillars of the consortium is to open clinical trials across all centres in collaboration with pharma companies, as exemplified by the Basket of Baskets (NCT03767075), Europe's largest precision oncology trial. Yet another example of successful collaboration between industry and non-for-profit institutions is CONNECT, the Norwegian Cancer Precision Medicine Implementation Consortium, launched in 2020 [108, 109]. The network now has 30 partners, including universities, companies and health organisations. Led by the Oslo Cancer Cluster, it addresses key obstacles in precision cancer medicine, fostering collaboration between public and private sectors. CONNECT drives a national framework for precision medicine, emphasising structured dialogues, workshops and international expertise. Operationalised through four working groups, it focuses on precision diagnostics, progress monitoring, funding pathways and data infrastructure on a Nordic level, showcasing effective academia-industry synergy.

Patient and next-of-kin collaboration

At the centre of personalised medicine is the patient. It is, therefore, necessary to involve patient representatives already in the planning stage of personalised oncology trials [110]. In a recent Swedish report focusing on developing patient and next-of-kin collaboration, a set of recommendations is provided for improved patient engagement [111]. For instance, it is recommended that the parties should, at an early stage, define common objectives and a clear process for evaluation, that all communication must be clear and transparent to avoid power imbalances, and that sustainable conditions for remuneration and representativity should be ensured, as well as access to information technology and management support. Finally, the report stresses the importance of sharing both positive and negative experiences beyond the partners to foster new forms of collaboration and as a basis for a continuous learning system.

In summary, patient representatives are today key stakeholders that should participate in all phases of oncology trials, a strategy adopted by an increasing number of clinical studies aiming at personalised medicine.

Final recommendations on molecular assays for study inclusion

In the last two decades, we have witnessed a major leap in the development of advanced diagnostic tools, which today allow us to provide personalised treatment and care. At the same time, these technologies can be used to identify patients suitable for inclusion and/or stratification in personalised oncology trials. Moreover, these tools enable further clinical, translational and basic research, thus not only providing clinical but also scientific, and through potential commercial exploitation, even economic benefit to society.

Based on our experience in developing and implementing personalised diagnostics in both routine practice and when planning a new personalised oncology trial, we recommend:

1. The use of comprehensive genomic profiling to identify patients for inclusion to personalised oncology trials. As a minimum, a large cancer gene panel, which also enables analysis of complex biomarkers, should be applied to analyse a tissue biopsy collected before study inclusion. In addition, the possibility to perform ctDNA analysis using a gene panel is recommended in patients where a tissue biopsy cannot be taken or was of insufficient quality.
2. The collection of additional material for exploratory biomarker analyses with emerging profiling technologies. Although validated gene panels are mostly used for study inclusion, the possibility of profiling the tumour with other assays, such as WES/WGS, WTS and proteomics, can provide additional insights associated with treatment outcomes. As the cost of these assays becomes more affordable and tissue handling is further developed, they will be available for up-front use.
3. The implementation of a CDSS tool to support clinical trial allocation and the establishment of a dedicated MTB. In this way, a more streamlined analysis of assay results can take place, which in turn provides harmonised treatment recommendations, especially in

studies opened across multiple centres. There should be a continuous development of the CDSS tool as a basis for a learning system in precision oncology. Additionally, it is of utmost importance to allow connection between trial networks, as personalised treatments are given to smaller patient populations.

4. The use of multi-modal diagnostics to take personalised oncology trials to the next level. By performing translational, retrospective research within trials, as well as designing appropriate observational studies, new diagnostic modalities and biomarkers can be tested before they can be implemented in future interventional trials. Coupling rich biomarker analysis data with available clinical data also allows for the development of machine learning and AI on a national and international level.
5. The involvement of patient representatives in all phases of personalised oncology trials from the study design to the final evaluation. The patient advocates are crucial for the dynamic implementation of new diagnostic modalities and therapeutic interventions in clinical trials.

These recommendations can hopefully provide guidance to study groups aiming at including high-throughput molecular assays as a basis for study inclusion. Nevertheless, as the field of personalised oncology is rapidly evolving, regular updates of the recommendations will be necessary.

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Conflict of interest statement

AE has received honoraria from Amgen, AstraZeneca, Bayer, Diaceutics, Pfizer and Roche. HGR has received research support from Illumina and NanoString and her institution has received honoraria for educational talks given by her (AstraZeneca, Pfizer, Merck, Roche, Illumina, Novartis, Incyte). JL has received honoraria from Roche, research support from AstraZeneca, Roche, Novartis, Ipsen and is shareholder on FenoMark Diagnostics Ab. DT has received honoraria from Roche, research support from Roche, Taiho oncology and Janssen. AS has received honoraria from Aignostics, Amgen, AstraZeneca, Astellas, Bayer, BMS, Eli Lilly, Illumina, Incyte, Janssen, MSD, Novartis, Pfizer, Qlucore, Roche, Seagen, Ervier, Takeda, and Thermo Fisher, and research grants from Bayer, BMS, Chugai, Incyte and MSD. RR has received honoraria from AbbVie, AstraZeneca, Janssen, Illumina, and Roche.

Data availability statement

Data sharing is not applicable to this article as no new data were created or analysed in this study.

References

1 Council of the European Union. Council conclusions on personalised medicine for patients. *OJC.* 2015;**C/421**:2–5.
2 Stenzinger A, Edsjö A, Ploeger C, Friedman M, Fröhling S, Wirta V, et al. Trailblazing precision medicine in Europe: a joint view by Genomic Medicine Sweden and the centers for personalized medicine, ZPM, in Germany. *Semin Cancer Biol.* 2022;**84**:242–54.
3 Edsjö A, Holmquist L, Geoerger B, Nowak F, Gomon G, Alix-Panabières C, et al. Precision cancer medicine: concepts, current practice, and future developments. *J Intern Med.* 2023;**294**:455–81.
4 Harris CA, Ward RL, Dobbins TA, Drew AK, Pearson S. The efficacy of HER2-targeted agents in metastatic breast cancer: a meta-analysis. *Ann Oncol.* 2011;**22**:1308–17.
5 Valachis A, Mauri D, Polyzos NP, Chlouverakis G, Mavroudis D, Georgoulias V. Trastuzumab combined to neoadjuvant chemotherapy in patients with HER2-positive breast cancer: a systematic review and meta-analysis. *Breast.* 2011;**20**:485–90.
6 Viani GA, Afonso SL, Stefano EJ, De Fendi LI, Soares FV. Adjuvant trastuzumab in the treatment of her-2-positive early breast cancer: a meta-analysis of published randomized trials. *BMC Cancer.* 2007;**7**:153.
7 Higuchi R, Dollinger G, Walsh PS, Griffith R. Simultaneous amplification and detection of specific DNA sequences. *Biotechnology.* 1992;**10**:413–17.
8 Higuchi R, Fockler C, Dollinger G, Watson R. Kinetic PCR analysis: real-time monitoring of DNA amplification reactions. *Biotechnology.* 1993;**11**:1026–30.
9 Nyren P, Pettersson B, Uhlen M. Solid phase DNA minisequencing by an enzymatic luminometric inorganic pyrophosphate detection assay. *Anal Biochem.* 1993;**208**:171–75.
10 Vogelstein B, Kinzler KW. Digital PCR. *Proc Natl Acad Sci USA.* 1999;**96**:9236–41.
11 Sanger F, Nicklen S, Coulson AR. DNA sequencing with chain-terminating inhibitors. *Proc Natl Acad Sci USA.* 1977;**74**:5463–67.
12 Heather JM, Chain B. The sequence of sequencers: the history of sequencing DNA. *Genomics.* 2016;**107**:1–8.
13 Fountzilas E, Tsimberidou AM, Vo HH, Kurzrock R. Clinical trial design in the era of precision medicine. *Genome Med.* 2022;**14**:101.
14 Mangat PK, Halabi S, Bruinooge SS, Garrett-Mayer E, Alva A, Janeway KA, et al. Rationale and design of the Targeted Agent and Profiling Utilization Registry (TAPUR) study. *JCO Precis Oncol.* 2018;**2**:1–14. doi: 10.1200/PO.18.00122
15 Van Der Velden DL, Hoes LR, Van Der Wijngaart H, Van Berge Henegouwen JM, Van Werkhoven E, Roepman P, et al. The drug rediscovery protocol facilitates the expanded use of existing anticancer drugs. *Nature.* 2019;**574**:127–31.
16 www.pcm4eu.eu
17 Sutton RT, Pincock D, Baumgart DC, Sadowski DC, Fedorak RN, Kroeker KI. An overview of clinical decision support systems: benefits, risks, and strategies for success. *NPJ Digit Med.* 2020;**3**:17.
18 Cheng DT, Mitchell TN, Zehir A, Shah RH, Benayed R, Syed A, et al. Memorial sloan kettering-integrated mutation profiling of actionable cancer targets (MSK-IMPACT): a hybridization capture-based next-generation sequencing clinical assay for solid tumor molecular oncology. *J Mol Diagn.* 2015;**17**:251–64.
19 Kazdal D, Hofman V, Christopoulos P, Ilié M, Stenzinger A, Hofman P. Fusion-positive non-small cell lung carcinoma: biological principles, clinical practice, and diagnostic implications. *Genes Chromosomes Cancer.* 2022;**61**:244–60.
20 Neumann O, Burn TC, Allgäuer M, Ball M, Kirchner M, Albrecht T, et al. Genomic architecture of FGFR2 fusions in cholangiocarcinoma and its implication for molecular testing. *Br J Cancer.* 2022;**127**:1540–49.
21 Amaral P, Carbonell-Sala S, De La Vega FM, Faial T, Frankish A, Gingeras T, et al. The status of the human gene catalogue. *Nature.* 2023;**622**:41–47.
22 Hutter C, Zenklusen JC. The cancer genome atlas: creating lasting value beyond its data. *Cell.* 2018;**173**:283–85.
23 Réda M, Richard C, Bertaut A, Niogret J, Collot T, Fumet JD, et al. Implementation and use of whole exome sequencing for metastatic solid cancer. *EBioMedicine.* 2020;**51**:102624.
24 Lier A, Penzel R, Heining C, Horak P, Fröhlich M, Uhrig S, et al. Validating comprehensive next-generation sequencing results for precision oncology: the NCT/DKTK molecularly aided stratification for tumor eradication research experience. *JCO Precis Oncol.* 2018;**2**:1–13.
25 Menzel M, Ossowski S, Kral S, Metzger P, Horak P, Marienfeld R, et al. Multicentric pilot study to standardize clinical whole exome sequencing (WES) for cancer patients. *NPJ Precis Oncol.* 2023;**7**:106.
26 Langenberg KPS, Meister MT, Bakhuizen JJ, Boer JM, Van Eijkelenburg NKA, Hulleman E, et al. Implementation of paediatric precision oncology into clinical practice: the individualized therapies for children with cancer program 'iTHER'. *Eur J Cancer.* 2022;**175**:311–25.
27 Cuppen E, Elemento O, Rosenquist R, Nikic S, Ijzerman M, Zaleski ID, et al. Implementation of whole-genome and transcriptome sequencing into clinical cancer care. *JCO Precis Oncol.* 2022;**6**:e2200245.
28 Jiao W, Atwal G, Polak P, Karlic R, Cuppen E, Al-Shahrour F, et al. A deep learning system accurately classifies primary and metastatic cancers using passenger mutation patterns. *Nat Commun.* 2020;**11**:728.
29 Schipper LJ, Samsom KG, Snaebjornsson P, Battaglia T, Bosch LJW, Lalezari F, et al. Complete genomic characterization in patients with cancer of unknown primary origin in routine diagnostics. *ESMO Open.* 2022;**7**:100611.
30 Meggendorfer M, Jobanputra V, Wrzeszczynski KO, Roepman P, De Bruijn E, Cuppen E, et al. Analytical demands to use whole-genome sequencing in precision oncology. *Semin Cancer Biol.* 2022;**84**:16–22.
31 Huang K-L, Mashl RJ, Wu Y, Ritter DI, Wang J, Oh C, et al. Pathogenic germline variants in 10,389 adult cancers. *Cell.* 2018;**173**:355–370.e14.
32 Jahn A, Rump A, Widmann TJ, Heining C, Horak P, Hutter B, et al. Comprehensive cancer predisposition testing within the prospective MASTER trial identifies hereditary cancer patients and supports treatment decisions for rare cancers. *Ann Oncol.* 2022;**33**:1186–99.

33 Horak P, Heining C, Kreutzfeldt S, Hutter B, Mock A, Hüllein J, et al. Comprehensive genomic and transcriptomic analysis for guiding therapeutic decisions in patients with rare cancers. *Cancer Discov.* 2021;**11**:2780–95.

34 Rosenquist R, Cuppen E, Buettner R, Caldas C, Dreau H, Elemento O, et al. Clinical utility of whole-genome sequencing in precision oncology. *Semin Cancer Biol.* 2022;**84**:32–39.

35 Sosinsky A, Ambrose J, Cross W, Turnbull C, Henderson S, Jones L, et al. Insights for precision oncology from the integration of genomic and clinical data of 13,880 tumors from the 100,000 Genomes Cancer Programme. *Nat Med.* 2024;**30**:279–89.

36 Stenzinger A, Moltzen EK, Winkler E, Molnar-Gabor F, Malek N, Costescu A, et al. Implementation of precision medicine in healthcare—a European perspective. *J Intern Med.* 2023;**294**:437–54.

37 Wadensten E, Wessman S, Abel F, Diaz De Ståhl T, Tesi B, Orsmark Pietras C, et al. Diagnostic yield from a nationwide implementation of precision medicine for all children with cancer. *JCO Precis Oncol.* 2023;**7**:e2300039.

38 Duncavage EJ, Schroeder MC, O'laughlin M, Wilson R, Macmillan S, Bohannon A, et al. Genome sequencing as an alternative to cytogenetic analysis in myeloid cancers. *N Engl J Med.* 2021;**384**:924–35.

39 Rezayee F, Eisfeldt J, Skaftason A, Öfverholm I, Sayyab S, Syvänen AC, et al. Feasibility to use whole-genome sequencing as a sole diagnostic method to detect genomic aberrations in pediatric B-cell acute lymphoblastic leukemia. *Front Oncol.* 2023;**13**:1217712.

40 Stacchiotti S, Maria Frezza A, Demetri GD, Blay J-Y, Bajpai J, Baldi GG, et al. Retrospective observational studies in ultra-rare sarcomas: a consensus paper from the Connective Tissue Oncology Society (CTOS) community of experts on the minimum requirements for the evaluation of activity of systemic treatments. *Cancer Treat Rev.* 2022;**110**:102455.

41 Samsom KG, Bosch LJW, Schipper LJ, Schout D, Roepman P, Boelens MC, et al. Optimized whole-genome sequencing workflow for tumor diagnostics in routine pathology practice. *Nat Protoc.* 2023;**19**(3):700–726.

42 Hoes LR, Van Berge Henegouwen JM, Van Der Wijngaart H, Zeverijn LJ, Van Der Velden DL, Van De Haar J, et al. Patients with Rare Cancers in the Drug Rediscovery Protocol (DRUP) benefit from genomics-guided treatment. *Clin Cancer Res.* 2022;**28**:1402–11.

43 André F, Rassy E, Marabelle A, Michiels S, Besse B. Forget lung, breast or prostate cancer: why tumour naming needs to change. *Nature.* 2024;**626**:26–29.

44 Chakraborty S, Martin A, Guan Z, Begg CB, Shen R. Mining mutation contexts across the cancer genome to map tumor site of origin. *Nat Commun.* 2021;**12**:3051.

45 Skaftason A, Qu Y, Abdulla M, Nordlund J, Berglund M, Ednersson SB, et al. Transcriptome sequencing of archived lymphoma specimens is feasible and clinically relevant using exome capture technology. *Genes Chromosom Cance.* 2022;**61**:27–36.

46 Lilljebjörn H, Orsmark-Pietras C, Mitelman F, Hagström-Andersson A, Fioretos T. Transcriptomics paving the way for improved diagnostics and precision medicine of acute leukemia. *Semin Cancer Biol.* 2022;**84**:40–49.

47 Perou CM, Sørlie T, Eisen MB, Van De Rijn M, Jeffrey SS, Rees CA, et al. Molecular portraits of human breast tumours. *Nature.* 2000;**406**:747–52.

48 Rosenwald A, Wright G, Chan WC, Connors JM, Campo E, Fisher RI, et al. The use of molecular profiling to predict survival after chemotherapy for diffuse large-B-cell lymphoma. *N Engl J Med.* 2002;**346**:1937–47.

49 Jensen M-B, Lænkholm A-V, Nielsen TO, Eriksen JO, Wehn P, Hood T, et al. The Prosigna gene expression assay and responsiveness to adjuvant cyclophosphamide-based chemotherapy in premenopausal high-risk patients with breast cancer. *Breast Cancer Res.* 2018;**20**:79.

50 Capper D, Jones DTW, Sill M, Hovestadt V, Schrimpf D, Sturm D, et al. DNA methylation-based classification of central nervous system tumours. *Nature.* 2018;**555**:469–74.

51 Pickles JC, Fairchild AR, Stone TJ, Brownlee L, Merve A, Yasin SA, et al. DNA methylation-based profiling for paediatric CNS tumour diagnosis and treatment: a population-based study. *Lancet Child Adolesc Health.* 2020;**4**:121–30.

52 Alexandrov LB, Kim J, Haradhvala NJ, Huang MN, Tian Ng AW, Wu Y, et al. Author correction: the repertoire of mutational signatures in human cancer. *Nature.* 2023;**614**:E41.

53 Koh G, Degasperi A, Zou X, Momen S, Nik-Zainal S. Mutational signatures: emerging concepts, caveats and clinical applications. *Nat Rev Cancer.* 2021;**21**:619–37.

54 Vikas P, Messersmith H, Compton C, Sholl L, Broaddus RR, Davis A, et al. Mismatch repair and microsatellite instability testing for immune checkpoint inhibitor therapy: ASCO endorsement of college of American pathologists guideline. *J Clin Oncol.* 2023;**41**:1943–48.

55 Alexandrov LB, Nik-Zainal S, Wedge DC, Aparicio SAJR, Behjati S, Biankin AV, et al. Signatures of mutational processes in human cancer. *Nature.* 2013;**500**:415–21.

56 Sha D, Jin Z, Budczies J, Kluck K, Stenzinger A, Sinicrope FA. Tumor mutational burden as a predictive biomarker in solid tumors. *Cancer Discov.* 2020;**10**:1808–25.

57 Miller RE, Leary A, Scott CL, Serra V, Lord CJ, Bowtell D, et al. ESMO recommendations on predictive biomarker testing for homologous recombination deficiency and PARP inhibitor benefit in ovarian cancer. *Ann Oncol.* 2020;**31**:1606–22.

58 Stewart MD, Merino Vega D, Arend RC, Baden JF, Barbash O, Beaubier N, et al. Homologous recombination deficiency: concepts, definitions, and assays. *Oncologist.* 2022;**27**:167–74.

59 Rempel E, Kluck K, Beck S, Ourailidis I, Kazdal D, Neumann O, et al. Pan-cancer analysis of genomic scar patterns caused by homologous repair deficiency (HRD). *NPJ Precis Oncol.* 2022;**6**:36.

60 Van Der Wijngaart H, Hoes LR, Van Berge Henegouwen JM, Van Der Velden DL, Zeverijn LJ, Roepman P, et al. Patients with biallelic BRCA1/2 inactivation respond to olaparib treatment across histologic tumor types. *Clin Cancer Res.* 2021;**27**:6106–14.

61 Mcgrail DJ, Pilié PG, Rashid NU, Voorwerk L, Slagter M, Kok M, et al. High tumor mutation burden fails to predict immune checkpoint blockade response across all cancer types. *Ann Oncol.* 2021;**32**:661–72.

62 Ignatiadis M, Sledge GW, Jeffrey SS. Liquid biopsy enters the clinic—implementation issues and future challenges. *Nat Rev Clin Oncol.* 2021;**18**:297–312.

63 Crowley E, Di Nicolantonio F, Loupakis F, Bardelli A. Liquid biopsy: monitoring cancer-genetics in the blood. *Nat Rev Clin Oncol.* 2013;**10**:472–84.
64 Chan KCA, Yeung S-W, Lui W-B, Rainer TH, Lo YMD. Effects of preanalytical factors on the molecular size of cell-free DNA in blood. *Clin Chem.* 2005;**51**:781–84.
65 Lampignano R, Neumann MHD, Weber S, Kloten V, Herdean A, Voss T, et al. Multicenter evaluation of circulating cell-free DNA extraction and downstream analyses for the development of standardized (pre)analytical work flows. *Clin Chem.* 2020;**66**:149–60.
66 Parpart-Li S, Bartlett B, Popoli M, Adleff V, Tucker L, Steinberg R, et al. The effect of preservative and temperature on the analysis of circulating tumor DNA. *Clin Cancer Res.* 2017;**23**:2471–77.
67 Barbany G, Arthur C, Liedén A, Nordenskjöld M, Rosenquist R, Tesi B, et al. Cell-free tumour DNA testing for early detection of cancer—a potential future tool. *J Intern Med.* 2019;**286**:118–36.
68 Edsjö A, Gisselsson D, Staaf J, Holmquist L, Fioretos T, Cavelier L, et al. Current and emerging sequencing-based tools for precision cancer medicine. *Mol Aspects Med.* 2024;**96**:101250.
69 Helland Å, Russnes HG, Fagereng GL, Al-Shibli K, Andersson Y, Berg T, et al. Improving public cancer care by implementing precision medicine in Norway: IMPRESS-Norway. *J Transl Med.* 2022;**20**:225.
70 Pascual J, Attard G, Bidard F-C, Curigliano G, De Mattos-Arruda L, Diehn M, et al. ESMO recommendations on the use of circulating tumour DNA assays for patients with cancer: a report from the ESMO Precision Medicine Working Group. *Ann Oncol.* 2022;**33**:750–68.
71 Wagner AH, Walsh B, Mayfield G, Tamborero D, Sonkin D, Krysiak K, et al. A harmonized meta-knowledgebase of clinical interpretations of somatic genomic variants in cancer. *Nat Genet.* 2020;**52**:448–57.
72 Chakravarty D, Gao J, Phillips SM, Kundra R, Zhang H, Wang J, et al. OncoKB: a precision oncology knowledge base. *JCO Precis Oncol.* 2017;**2017**(1):PO1700011.
73 Suehnholz SP, Nissan MH, Zhang H, Kundra R, Nandakumar S, Lu C, et al. Quantifying the expanding landscape of clinical actionability for patients with cancer. *Cancer Discov.* 2024;**14**:49–65.
74 International Cancer Genome C, Hudson TJ, Anderson W, Artez A, Barker AD, Bell C, et al. International network of cancer genome projects. *Nature.* 2010;**464**:993–98.
75 https://www.cancer.gov/tcga
76 Sanchez-Vega F, Mina M, Armenia J, Chatila WK, Luna A, La KC, et al. Oncogenic signaling pathways in the cancer genome atlas. *Cell.* 2018;**173**:321–337.e10.
77 Bakhtiar H, Helzer KT, Park Y, Chen Yi, Rydzewski NR, Bootsma ML, et al. Identification of phenocopies improves prediction of targeted therapy response over DNA mutations alone. *NPJ Genom Med.* 2022;**7**:58.
78 Lei JT, Zhang B. Proteogenomics drives therapeutic hypothesis generation for precision oncology. *Br J Cancer.* 2021;**125**:1–3.
79 Boys EL, Liu J, Robinson PJ, Reddel RR. Clinical applications of mass spectrometry-based proteomics in cancer: where are we? *Proteomics.* 2023;**23**:e2200238.
80 Herbst SA, Vesterlund M, Helmboldt AJ, Jafari R, Siavelis I, Stahl M, et al. Proteogenomics refines the molecular classification of chronic lymphocytic leukemia. *Nat Commun.* 2022;**13**:6226.
81 Zhang B, Whiteaker JR, Hoofnagle AN, Baird GS, Rodland KD, Paulovich AG. Clinical potential of mass spectrometry-based proteogenomics. *Nat Rev Clin Oncol.* 2019;**16**:256–68.
82 Zhu Y, Orre LM, Johansson HJ, Huss M, Boekel J, Vesterlund M, et al. Discovery of coding regions in the human genome by integrated proteogenomics analysis workflow. *Nat Commun.* 2018;**9**:903.
83 Sun BB, Chiou J, Traylor M, Benner C, Hsu Yi-H, Richardson TG, et al. Plasma proteomic associations with genetics and health in the UK Biobank. *Nature.* 2023;**622**:329–38.
84 Lewis SM, Asselin-Labat M-L, Nguyen Q, Berthelet J, Tan X, Wimmer VC, et al. Spatial omics and multiplexed imaging to explore cancer biology. *Nat Methods.* 2021;**18**:997–1012.
85 Askin S, Burkhalter D, Calado G, El Dakrouni S. Artificial intelligence applied to clinical trials: opportunities and challenges. *Health Technol.* 2023;**13**:203–13.
86 Lehtiö J, Arslan T, Siavelis I, Pan Y, Socciarelli F, Berkovska O, et al. Proteogenomics of non-small cell lung cancer reveals molecular subtypes associated with specific therapeutic targets and immune evasion mechanisms. *Nat Cancer.* 2021;**2**:1224–42.
87 Lang F, Schrörs B, Löwer M, Türeci Ö, Sahin U. Identification of neoantigens for individualized therapeutic cancer vaccines. *Nat Rev Drug Discov.* 2022;**21**:261–82.
88 Fournier L, Costaridou L, Bidaut L, Michoux N, Lecouvet FE, De Geus-Oei L-F, et al. Incorporating radiomics into clinical trials: expert consensus endorsed by the European Society of Radiology on considerations for data-driven compared to biologically driven quantitative biomarkers. *Eur Radiol.* 2021;**31**:6001–12.
89 Cobo M, Menéndez Fernández-Miranda P, Bastarrika G, Lloret Iglesias L. Enhancing radiomics and deep learning systems through the standardization of medical imaging workflows. *Sci Data.* 2023;**10**:732.
90 Napoli GC, Figg WD, Chau CH. Functional drug screening in the era of precision medicine. *Front Med.* 2022;**9**:912641.
91 Horak P, Griffith M, Danos AM, Pitel BA, Madhavan S, Liu X, et al. Standards for the classification of pathogenicity of somatic variants in cancer (oncogenicity): joint recommendations of Clinical Genome Resource (ClinGen), Cancer Genomics Consortium (CGC), and Variant Interpretation for Cancer Consortium (VICC). *Genet Med.* 2022;**24**:1991.
92 Mateo J, Chakravarty D, Dienstmann R, Jezdic S, Gonzalez-Perez A, Lopez-Bigas N, et al. A framework to rank genomic alterations as targets for cancer precision medicine: the ESMO Scale for clinical actionability of molecular targets (ESCAT). *Ann Oncol.* 2018;**29**:1895–902.
93 Nakken S, Fournous G, Vodák D, Aasheim LB, Myklebost O, Hovig E. Personal Cancer Genome Reporter: variant interpretation report for precision oncology. *Bioinformatics.* 2018;**34**:1778–80.
94 Tamborero D, Dienstmann R, Rachid MH, Boekel J, Baird R, Braña I, et al. Support systems to guide clinical decision-making in precision oncology: the Cancer Core Europe Molecular Tumor Board Portal. *Nat Med.* 2020;**26**:992–94.
95 Landrum MJ, Chitipiralla S, Brown GR, Chen C, Gu B, Hart J, et al. ClinVar: improvements to accessing data. *Nucleic Acids Res.* 2020;**48**:D835–44.

96 Krysiak K, Danos AM, Saliba J, Mcmichael JF, Coffman AC, Kiwala S, et al. CIViCdb 2022: evolution of an open-access cancer variant interpretation knowledgebase. *Nucleic Acids Res.* 2023;**51**:D1230–41.
97 Patterson SE, Statz CM, Yin T, Mockus SM. Utility of the JAX Clinical Knowledgebase in capture and assessment of complex genomic cancer data. *NPJ Precis Oncol.* 2019;**3**:2.
98 Tamborero D, Dienstmann R, Rachid MH, Boekel J, Lopez-Fernandez A, Jonsson M, et al. The Molecular Tumor Board Portal supports clinical decisions and automated reporting for precision oncology. *Nat Cancer.* 2022;**3**:251–61.
99 Eggermont AMM, Apolone G, Baumann M, Caldas C, Celis JE, De Lorenzo F, et al. Cancer core Europe: A translational research infrastructure for a European mission on cancer. *Mol Oncol.* 2019;**13**:521–27.
100 https://eosc4cancer.eu/
101 Oehring R, Ramasetti N, Ng S, Roller R, Thomas P, Winter A, et al. Use and accuracy of decision support systems using artificial intelligence for tumor diseases: a systematic review and meta-analysis. *Front Oncol.* 2023;**13**:1224347.
102 Bot N, Waelli M. Implementing a clinical cutting-edge and decision-making activity: an ethnographic teamwork approach to a molecular tumorboard. *BMC Health Serv Res.* 2020;**20**:922.
103 Rao S, Pitel B, Wagner AH, Boca SM, Mccoy M, King I, et al. Collaborative, multidisciplinary evaluation of cancer variants through virtual molecular tumor boards informs local clinical practices. *JCO Clin Cancer Inform.* 2020;**4**:602–13.
104 Gray SW, Hicks-Courant K, Cronin A, Rollins BJ, Weeks JC. Physicians' attitudes about multiplex tumor genomic testing. *J Clin Oncol.* 2014;**32**:1317–23.
105 Dutta R, Vallurupalli M, Mcveigh Q, Huang FW, Rebbeck TR. Understanding inequities in precision oncology diagnostics. *Nat Cancer.* 2023;**4**:787–94.
106 Kahles A, Goldschmid H, Volckmar A-L, Ploeger C, Kazdal D, Penzel R, et al. Regulation (EU) 2017/746 (IVDR): practical implementation of annex I in pathology. *Pathologie (Heidelb).* 2023;**44**:86–95.
107 Kahles A, Goldschmid H, Volckmar A-L, Ploeger C, Kazdal D, Penzel R, et al. Structure and content of the EU-IVDR: current status and implications for pathology. *Pathologie (Heidelb).* 2023;**44**:73–85.
108 Taskén K, Russnes HEG, Aas E, Bjørge L, Blix ES, Ahlquist TC, et al. A national precision cancer medicine implementation initiative for Norway. *Nat Med.* 2022;**28**:885–87.
109 www.connectnorway.org/
110 Geissler J, Ryll B, Di Priolo SL, Uhlenhopp M. Improving patient involvement in medicines research and development: a practical roadmap. *Ther Innov Regul Sci.* 2017;**51**:612–19.
111 https://biobanksverige.se/wp-content/uploads/Patient-and-next-of-kin-collaboration-for-better-research-and-healthcare-2-0.pdf

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