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Review article

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Rare molecular subtypes of lung cancer

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Abstract

Oncogenes that occur in ≤5% of non-small-cell lung cancers have been defined as 'rare'; nonetheless, this frequency can correspond to a substantial number of patients diagnosed annually. Within rare oncogenes, less commonly identified alterations (such as *HRAS*, *NRAS*, *RIT1*, *ARAF*, *RAF1* and *MAP2K1* mutations, or ERBB family, *LTK* and *RASGRF1* fusions) can share certain structural or oncogenic features with more commonly recognized alterations (such as *KRAS*, *BRAF*, *MET* and ERBB family mutations, or *ALK*, *RET* and *ROS1* fusions). Over the past 5 years, a surge in the identification of rare-oncogene-driven lung cancers has challenged the boundaries of traditional clinical grade diagnostic assays and profiling algorithms. In tandem, the number of approved targeted therapies for patients with rare molecular subtypes of lung cancer has risen dramatically. Rational drug design has iteratively improved the quality of small-molecule therapeutic agents and introduced a wave of antibody-based therapeutics, expanding the list of actionable de novo and resistance alterations in lung cancer. Getting additional molecularly tailored therapeutics approved for rare-oncogene-driven lung cancers in a larger range of countries will require ongoing stakeholder cooperation. Patient advocates, health-care agencies, investigators and companies with an interest in diagnostics, therapeutics and real-world evidence have already taken steps to surmount the challenges associated with research into low-frequency drivers.

Sections

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Key points

- Many 'rare' molecular subtypes of lung cancer individually account for a substantial number of patients diagnosed annually around the world.
- An incredible diversity of molecular subtypes of lung cancer exists; mechanistically, these can be classified into mutations, fusions and copy number changes.
- Alterations involving receptor tyrosine kinases and MAPK pathway members can share structural and/or oncogenic features; conversely, other alterations have distinct mechanisms of oncogenesis such as effects on RNA splicing or epigenetic processes.
- Optimizing the identification of rare driver oncogenes requires both clinicopathological approaches that are feature-agnostic and tailored approaches to patient selection, tumour and plasma interrogation, DNA and RNA sequencing, and more unbiased profiling.
- Targeted therapy approvals were previously focused on certain alterations to the point of saturation and dominated by small-molecule tyrosine kinase inhibitors; approved and investigational antibody-based therapies are now becoming more widely used.
- Oncogene-driven advocacy, the adoption of contemporary trial designs, expedited regulatory pathways for drug development, and real-world evidence generation are all crucial steps towards promoting research and expediting the approval of drugs for rare oncogene-driven lung cancers.

Introduction

No unified definition of a rare molecular subtype of lung cancer exists[1]. In Europe and Asia, a cancer is considered rare if it occurs in <6 per 100,000 people annually[2,3]. In the USA, a cancer is rare if it occurs in <15 per 100,000 people annually[4]. On top of this, molecular subsets of lung cancer have also been classified as rare in the literature not by annual incidence, but by percentage frequency[5] (for example, ≤5% of non-small-cell lung cancers (NSCLCs)) (Supplementary Table 1). Fortunately, molecular subsets of lung cancer that meet the ≤5% cut-off are expected to constitute ≤2 cases per 100,000 people annually, thus also satisfying the definitions of a rare cancer applied across Europe, Asia and the USA.

Many rare molecular subtypes of lung cancer exist (Fig. 1a), recognizing that frequency might also vary by race and/or ethnicity (Fig. 1b, Supplementary Table 2), age (Supplementary Fig. 1) and the detection assay used (Fig. 1c). Importantly, categorizing these cancers as rare hinders the appreciation of their true incident burden; some of these subtypes might affect 18,000 to >90,000 people annually worldwide[6,7] (Supplementary Table 3). Furthermore, selected rare-oncogene-driven NSCLCs are diagnosed annually with a frequency comparable to or even exceeding that of several other malignancies (such as acute lymphocytic leukaemias, and vulvar, bone and male genital cancers)[8,9] (Fig. 1d).

In this Review, we summarize the molecular, clinical and pathological features of rare molecular subtypes of lung cancer, focusing on likely oncogenic alterations that tend to be mutually exclusive with other bona fide drivers. Rare molecular subtypes of lung cancer fall into two major categories. The first category is composed of more commonly recognized subtypes such as those with mutations in *EGFR* exon 20, *ERBB2*, *BRAF*^{V600E} and *MET*, and *ALK*, *ROS1*, *RET* and *NTRK1/2/3* fusion-positive lung cancers. The second category is composed of cancers harbouring less commonly recognized alterations (such as *KRAS*, *HRAS*, *NRAS*, *DDR2*, *LTK*, *RIT1*, *ARAF*, non-V600E *BRAF*, *RAF1* and *MAP2K1* alterations) for which data are much more limited.

The molecular features, oncogenesis and signalling pathways, and clinicopathological features of the less-recognized lung cancer subtypes are often grouped with those of more commonly recognized subtypes to underscore similarities and differences. For example, features of alterations in genes encoding MAPK family members, such as *HRAS*, *NRAS* and *RIT1* mutations, are discussed alongside *KRAS* mutations.

The same approach is taken in discussing diagnosis and treatment. Diagnostic strategies to maximize *RET* and *ROS1* fusion detection, for instance, apply to *RASGRF1* fusions. While many commonly recognized subtypes are considered highly actionable owing to the availability of an approved targeted therapy in one or more regulatory environments, less-common subtypes might also be considered actionable based on varying levels of preclinical and clinical evidence (Supplementary Table 4). An example is the preclinical activity of tyrosine kinase inhibitors (TKIs) used to treat ALK fusions in lung cancer models with LTK fusions.

Finally, efforts to expedite targeted therapy approvals for rare molecular subtypes of lung cancer are outlined. We discuss how multiple stakeholders have come together to increase the ease with which data on the activity of such therapies are generated, and how these initiatives are challenging the established models for regulatory approval that historically have not favoured low-frequency alterations.

Molecular subtypes

Molecular features

Mutations. Rare mutations or sequence variants can be classified in terms of the proteins encoded by the affected genes. One group comprises mutations in the genes encoding receptor tyrosine kinases (RTKs) (Fig. 2), including genes such as *MET*, *EGFR*, *ERBB2* and *DDR2*. A spectrum of mutations can affect these genes, although the most common mutations are *MET* exon 14 alterations (4% of NSCLCs[10]), *EGFR* exon 20 mutations (1.5% of NSCLCs[11]), *ERBB2* exon 20 mutations (1.4% of NSCLCs[11]) and *DDR2* mutations (4% of squamous lung cancers[12] and 0.4% of lung adenocarcinomas (LUADs)[13]).

Mutations in genes encoding members of the MAPK signalling pathway are another group (Fig. 3). These mutations involve genes such as *KRAS*, *NRAS*, *HRAS*, *RIT1*, *ARAF*, *BRAF*, *RAF1* and *MAP2K1*. Mutations that affect RAS protein family members include *KRAS* (non-G12C/V/D mutations individually occur in <5% of NSCLCs[14]), *NRAS* (0.9% of NSCLCs), *HRAS* (0.1% of NSCLCs) and *RIT1* (~0.7% of LUADs). *KRAS* G12C, G12V and G12D mutations exceed the frequency threshold for rare molecular subtypes and are excluded from this discussion. Mutations that affect downstream signalling proteins in LUADs involve *ARAF* (0.2%), *BRAF* (4.5%), *RAF1* (0.4%) and *MAP2K1* (0.7%) (Supplementary Table 2).

Rare mutations can also be classified by alteration type. Missense point mutations that result in amino acid substitutions can be found in *KRAS*, *NRAS*, *HRAS*, *RIT1*, *ARAF*, *BRAF*, *RAF1*, *MAP2K1* and *DDR2*. Insertions and/or deletions (indels) can affect *EGFR*, *ERBB2* and *MET*. *EGFR* and *ERBB2* exon 20 insertions are structurally paralogous. Kinase domain duplications of ERBB family members (0.2% of NSCLCs[15]), such as in-tandem and in-frame duplications of exons 18–25 in *EGFR*, can

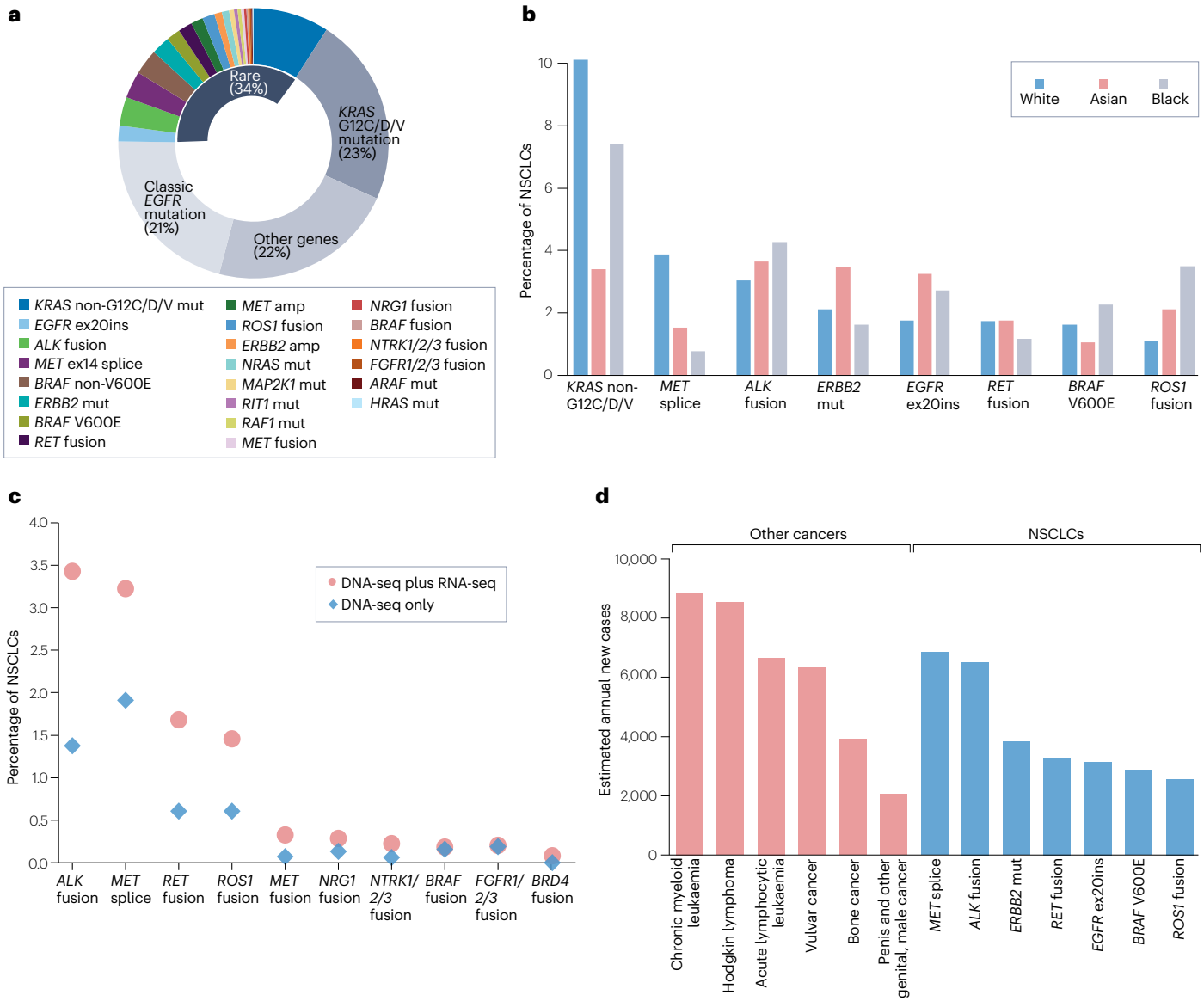


Fig. 1 | Frequency of 'rare' lung cancers. The prevalence of oncogenic driver alterations in lung adenocarcinomas was based on data from two aggregated cohorts. The first cohort, used to calculate the prevalence of non-fusion and non-*MET* exon 14 alterations, was derived from the GENIE database (v12; $n = 16{,}913$)[13], the PanCancer Atlas cohort of The Cancer Genome Atlas (TCGA; $n = 566$)[193] and the OncoSG cohort ($n = 305$)[194]. The prevalence of fusions and *MET* exon 14 alterations was based on data from cohorts with both DNA sequencing (DNA-seq) and RNA sequencing (RNA-seq) results available: MSK-IMPACT 468 and 505 (Genie v.12.0)[13], TCGA[193] and OncoSG[194]. All data were extracted from and visualized using cBioPortal[195,196]. **a**, The prevalence of oncogene-driven lung adenocarcinomas is shown. Rare lung cancers comprise over a third of cases. 'Other genes' comprises alterations that are not actionable or have yet undefined oncogenicity; no known mitogenic drivers were identified in this group. The *KRAS* non-G12C/D/V group comprises mutations that individually are present in <5% of non-small-cell lung cancers (NSCLCs). **b**, Prevalence of an alteration can vary by race and/or ethnicity. Using the same dataset, the prevalence of oncogenic drivers in white, Asian and Black patients is shown. **c**, Prevalence can also vary by the type of assay used. A comparison of the prevalence of fusions and *MET* exon 14 alterations between panels using DNA-seq only (all panels at Genie v.12.0 except MSK IMPACT 468 and 505 (ref. [13])) and panels including both DNA-seq and RNA-seq (MSK-IMPACT 468 and 505 (ref. [13]), TCGA[193] and OncoSG[194]) is shown. **d**, Estimates of the annual incidence of each molecular subtype of NSCLC in the USA are derived from the GENIE database (v12; $n = 19{,}777$ NSCLCs[13]) and statistics from Cancer.Net[197] that summarized data from the American Cancer Society, Cancer Facts & Figures 2022, and the International Agency for Research on Cancer. amp, amplification; mut, mutation.

also occur. Notably, kinase domain duplications have been observed in other, non-ERBB genes (such as *RET* and *MET*). For *MET* exon 14 alterations, these include indels involving the splice sites flanking exon 14.

Notably, while certain alteration types are more commonly associated with particular genes, other alterations may be observed as well in these same genes. Point mutations are more common in *KRAS*, *RIT1*,

BRAF and *MAP2K1*, although indels can also be identified. Conversely, beyond the more common indels, point mutations are sometimes identified in *MET* (kinase or semaphorin domains) and *ERBB2* (kinase, transmembrane or extracellular domains)[13].

Fusions. Fusions can be classified based on the proteins encoded by the affected genes (Figs. 4 and 5). One group involves genes encoding RTKs, including *ALK* (3–4% of LUADs), *RET* (1–2% of LUADs), *ROS1* (1–2% of LUADs), *NTRK1/2/3* (<1% of LUADs), *FGFR1/2/3* (<1% of LUADs), *EGFR* (<1% of LUADs), *ERBB2* (<1% of LUADs), *ERBB4* (<1% of LUADs) and *LTK* (<1% of LUADs) fusions. These oncogenic fusions typically include an intact kinase domain. A second group involves genes encoding members of the MAPK signalling pathway. These include *RASGRF1* (<0.1% of LUADs)[16] and *BRAF* (0.2% of LUADs)[17] fusions (Supplementary Table 2). *BRAF*-containing fusions generally include the kinase domain and are structurally similar to RTK fusions, whereas *RASGRF1*-containing fusions include the catalytically active carboxy-terminal RAS–GEF domain of RASGRF1 (ref. [16]). Another group involves genes encoding RTK ligands including *NRG1* (0.3% of LUADs) and *NRG2* fusions (0.02% of NSCLCs)[18,19]. Other fusions that do not belong in any one of these

Fig. 2 | Receptor tyrosine kinase gene mutations. a, Mutations involving *EGFR*, *ERBB2* and *DDR2* that result in the expression of putatively ligand-independent constitutively active oncogenic kinases are shown. *EGFR* insertions (such as the 9-bp insertions SVD or NPH, or the 12-bp insertion FQEA) typically occur between residues 769 and 775 while *ERBB2* insertions (such as the 12-bp insertion YVMA) occur between residues 775 and 881. *DDR2* mutations can affect both extracellular domains (such as *DDR2* G253S) and intracellular domains (such as *DDR2* G774V). **b**, *MET* mutations can similarly occur in various extracellular and intracellular domains. *MET* exon 14 alterations are thought to be ligand-dependent and therefore are reliant on the endogenous ligand hepatocyte growth factor (HGF). These mutation types are not intuitively annotated in some cases and can often be missed by clinicians when reviewing reports, particularly when the variants lie deep within introns and their effect on splicing remains unclear. The *MET* exon 14 alteration c.2888-40_2888-19del20 provides an example of this scenario. This alteration is a 20-bp deletion located in the intronic region adjacent to the splice acceptor site at the 5′ boundary of *MET* exon 14; the deletion involves positions −40 to −19 preceding the start of the exon 14 coding sequence at position 2888. RNA sequencing may be helpful to confirm *MET* exon 14 skipping in these cases with deep intronic mutations. The variants were selected from lung adenocarcinomas in the GENIE database (v12)[13] and reflect driver mutations as annotated using OncoKB and hotspot recurrence[198]. TK, tyrosine kinase.

three categories include those involving *BRD4* (0.05% of NSCLCs[20]) and *PKC*. *PKC* fusions in particular should be considered a separate entity as these are loss-of-function alterations[21].

A wide variety of kinase fusion partners exist. Some partners predominantly fuse with a specific RTK (such as *EML4* with *ALK*[22]); others, such as members of the TRIM protein family, can fuse with more than one RTK (such as *TRIM24-RET* and *TRIM24-NTRK2*)[23]. Partners may influence the localization of the kinase, including to transmembrane regions (such as *NRG1* or *RASGRF1* fusions) or subcellular locations (such as *NTRK1/2/3* fusions)[24]. Fusion partners can likewise affect the ability to undergo ligand-independent dimerization by contributing additional dimerization domains (including coiled-coil, zinc finger, LisH, WDR or SAM domains)[23].

Copy number alterations. Amplifications of the RTK-encoding genes *ERBB2* and *MET* occur in 0.9% and 1.4% of patients with newly diagnosed LUADs, respectively (Supplementary Table 2). *FGFR1* and *ARAF* amplifications have been identified in 1–3% and 1% of such patients, respectively[25,26]. Other less-well-characterized copy-number alterations include *EGFR*, *BRAF* and *KRAS* amplifications[8,13].

Amplifications can occur in chromosomal or extrachromosomal DNA[27] (double minutes) and are also found as mechanisms of secondary resistance to EGFR TKIs in patients with *EGFR*-mutant lung cancers[28]. Higher levels of amplification and focality can correlate with increased dependence on the amplified gene[29].

Oncogenesis and signalling

Levels of evidence supporting a role of rare molecular alterations or sequence variants in lung cancer oncogenesis vary considerably depending on the alteration. These range from alterations with compelling evidence of oncogenicity, such as *MET* exon 14 skipping mutations, to alterations requiring further investigation, such as fusions involving *BRD4*.

RTK and RTK ligand alterations. Mutations, fusions and amplifications involving genes encoding RTKs and their ligands functionally converge on increased RTK activity and the activation of downstream signalling pathways; these preferentially include the MAPK, PI3K, PKC and JAK–STAT signalling pathways. Increased RTK activity can occur in either a ligand-dependent or ligand-independent manner.

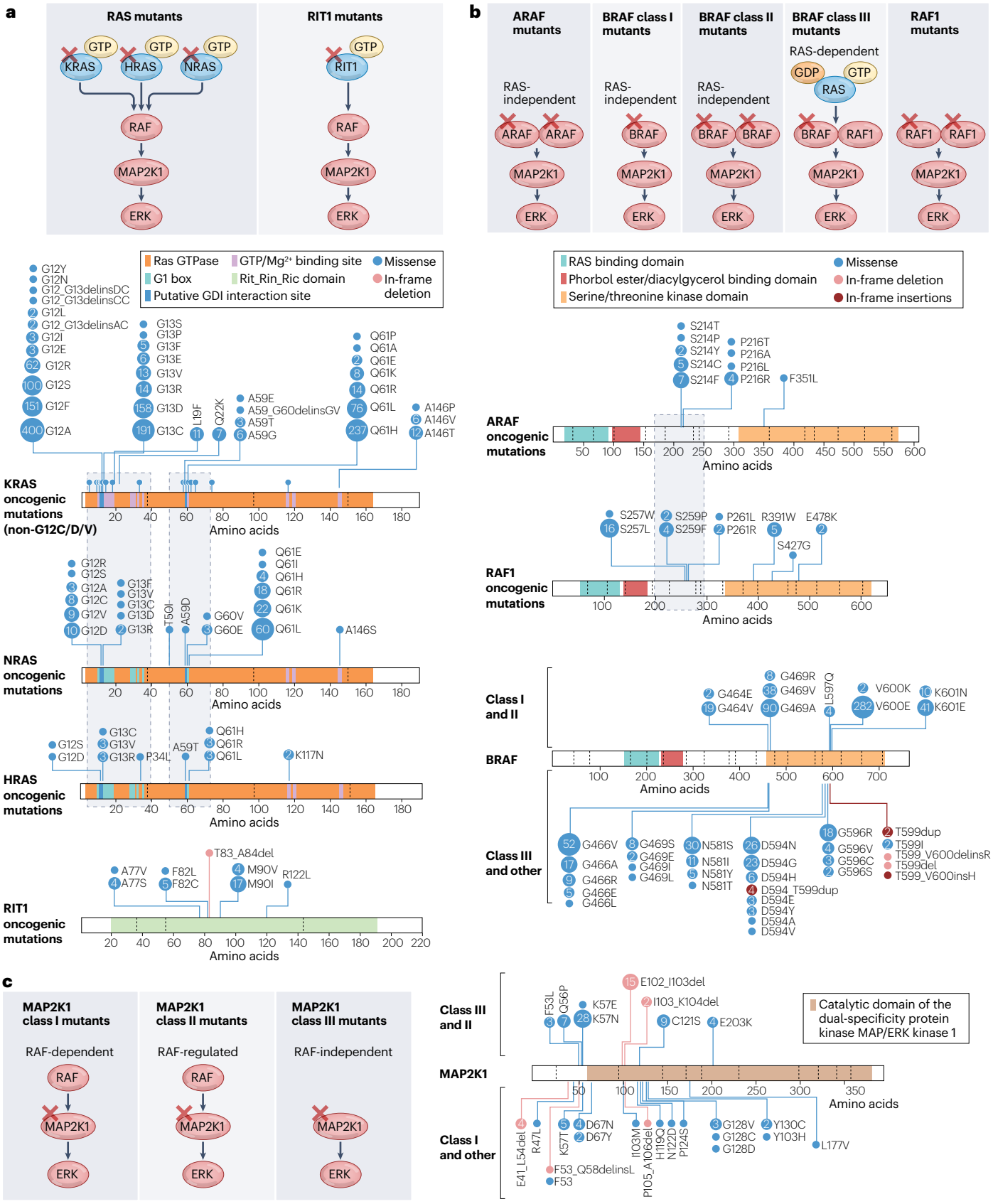
Ligand-independent constitutive kinase domain activation can occur with mutations or fusions involving *EGFR*, *ERBB2*, *ERBB4*, *DDR2*, *ALK*, *RET*, *ROS1*, *NTRK1/2/3* and *LTK*[12,30,31]. Such mutant RTKs maintain their transmembrane localization. By contrast, although certain RTK fusions are known to localize to the cell membrane, many localize to the cytoplasm or other subcellular compartments (Fig. 5e). Localization differences can modify the activation of downstream signalling pathways[32].

Ligand-dependent RTK activation occurs with altered splicing. Many *MET* exon 14 alterations interfere with splice acceptor and/or donor sites, leading to exon 14 skipping. Without the CBL ubiquitin ligase binding domain encoded by exon 14, MET is recycled to the cell surface rather than degraded[33]. *ERBB2* exon 16 skipping mutations (*ERBB2*$^{\Delta ex16}$) and *FGFR2* exon 18 truncated alterations (*FGFR2*$^{\Delta ex18}$) have also been identified[34,35]. These mutations eliminate the regulatory elements of HER2 and FGFR2, respectively, and induce constitutive receptor dimerization. Ligand-dependent RTK activation also occurs with amplification of several genes encoding RTKs (such as *EGFR*, *ERBB2*, *MET*, *FGFR1* and *FGFR3*). Such amplifications can increase the cell surface density of RTKs that remain influenced by ligand binding. Higher levels of amplification might correspond with higher RTK levels[29].

Fusions that involve *NRG1* or *NRG2* produce chimeric oncoproteins that retain an EGF-like domain, which binds HER3 and HER4 in an autocrine or paracrine fashion. While >30 different *NRG1* isoforms exist, NRG1-containing fusions preferentially occur with the NRG1 IIIβ-isoform, which is known to have a higher affinity for HER3 and HER4 than the α-isoform. Receptor dimerization (such as HER2 with HER3) then occurs, leading to activation of the MAPK, PI3K and FAK signalling pathways[18].

MAPK pathway alterations. RAS proteins are GTPases with biological activity governed by nucleotide binding states. The ratio of inactive RAS–GDP to active RAS–GTP is determined by the relative rates of GDP to GTP exchange and GTP hydrolysis. *KRAS*, *NRAS* and *HRAS* mutations, and *RASGRF1* fusions, can influence either or both of these activities[36]. Most *KRAS*, *NRAS* and *HRAS* codon 12 mutants affect GTP hydrolysis without changing the rate of GDP-to-GTP exchange, while codon 13 mutants affect both activities. The C-terminal domain of *RASGRF1* fusions catalyses the dissociation of GDP from RAS proteins[16]. Inactivating mutations in *NF1* and *RASA1* have also been identified; both genes encode RAS GTPase-activating proteins that negatively regulate the RAS signalling pathway[37]. Finally, *RIT1* mutations have been shown to activate MAPK signalling by escaping LZTR1-mediated degradation[38].

Mutations in genes encoding members of the RAF family can be grouped by RAS dependency. RAS-independent activation of MAP2K1 and MAP2K2 occurs with *ARAF* S214X, *BRAF* class I (such as V600E) and II mutations (such as G469A and K601E), and *RAF1* mutations. *BRAF* class I mutations signal as monomers. *BRAF* fusions and *ARAF*, *BRAF* class II and *RAF1* mutations signal as dimers[26,39,40]. RAS-dependent activation occurs with *BRAF* class III mutations (such as G466V, D594G and N581S), which have either impaired kinase activity or are kinase-dead and have a greater affinity for RAS–GTP than wild-type BRAF. RAS–GTP


a
RAS mutants
RIT1 mutants
GTP
KRAS
HRAS
NRAS
RIT1
RAF
MAP2K1
ERK
b
ARAF mutants
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BRAF class III mutants
RAF1 mutants
RAS-independent
RAS-dependent
GDP
RAS
ARAF
BRAF
RAF1
Ras GTPase
G1 box
Putative GDI interaction site
GTP/Mg2+ binding site
Rit_Rin_Ric domain
Missense
In-frame deletion
RAS binding domain
Phorbol ester/diacylglycerol binding domain
Serine/threonine kinase domain
In-frame insertions
KRAS oncogenic mutations (non-G12C/D/V)
NRAS oncogenic mutations
HRAS oncogenic mutations
RIT1 oncogenic mutations
ARAF oncogenic mutations
RAF1 oncogenic mutations
Class I and II
BRAF
Class III and other
Amino acids
c
MAP2K1 class I mutants
MAP2K1 class II mutants
MAP2K1 class III mutants
RAF-dependent
RAF-regulated
RAF-independent
Class III and II
MAP2K1
Class I and other
Catalytic domain of the dual-specificity protein kinase MAP/ERK kinase 1

Fig. 3 | RAS and MAPK family mutations. a, Mutations in *KRAS*, *NRAS* and *HRAS* can affect a variety of residues, including the paralogous (blue rectangles with dashed borders) G12, G13 and Q61 residues. *RIT1* encodes a small GTPase. *RIT1* mutations can affect the A77, F82 and M90 residues located close to the switch II pocket. **b**, *ARAF* mutations commonly affect S214 in addition to other residues. To date, mutations in the S214 codons are the only mutations in *ARAF* that are proven to be oncogenic in patients with lung cancer. *RAF1* mutations involve S257 and S259 (an *ARAF* S214 paralogue). *BRAF* mutations affect V600 and a wide variety of non-V600 residues (such as G466, G469, N581, D594, G596 and K601) located in the serine–threonine kinase domain. *BRAF* V600E is considered a class I (RAS-independent) alteration, while class II (RAS-independent) and class III (RAS-dependent) alterations comprise many non-V600 substitutions. **c**, *MAP2K1* class I mutations (such as D67N) are RAF-dependent. Class II mutations (such as K57N, a common *MAP2K1* mutation) can be modulated when phosphorylated by RAF and can occur either in isolation or co-occur with ERK-activating alterations. Class III mutations are both RAF and phosphorylation-independent, constitutively active and highly oncogenic. The variants were selected from lung adenocarcinomas in the GENIE database (v12)[13] and reflect driver mutations as annotated by OncoKB and hotspot recurrence[198].

binding of these class III mutant proteins results in enhanced RAF1 activity and increased ERK signalling[39] and other RAS signalling pathway alterations might co-occur. Similarly, *ARAF* amplifications can activate RAS in a kinase-independent manner by antagonizing NF1 binding[26].

MAP2K1 mutations can be grouped by RAF dependency. *MAP2K1* class I mutations (such as D67N and P124S) are RAF-dependent, have a limited transforming capacity and can co-occur alongside other ERK-activating alterations. *MAP2K1* class II (such as K57N and C121S) and class III alterations (such as E102_I103del and I103-K104del) are RAF-independent[41].

Other alterations. BRD4 fusions are well described in the context of NUT midline carcinomas. In lung cancer, fusions such as BRD4–NOTCH3 (Fig. 5d) might sequester histone acetyltransferases and other transcriptional co-factors to chromatin regions that transcribe selected genes (including *MYC*)[42]. Notably, this fusion includes the functional ankyrin domain of NOTCH3, and such fusions have been described as constitutive activators of NOTCH signalling in other tumours[43]. Therefore, both BRD4 and NOTCH3 domains are likely to contribute to the oncogenicity of BRD4–NOTCH3.

Clinicopathological characteristics

Clinical and histological features. Most rare driver alterations in RTKs, RAS, RAF and MEK are enriched in LUADs (Supplementary Table 5), the most common histological subtype of NSCLC[8,44]. These alterations can also be found in non-LUAD histologies such as squamous cell, large cell neuroendocrine, or rarely small-cell lung cancers. For example, *DDR2* mutations and *FGFR1* amplifications are enriched in squamous cell carcinomas, a histological subtype that also preferentially harbours *PIK3CA* alterations[45]. No pathological feature is entirely specific for a given molecular driver, although certain unique morphological patterns are associated with rare genomic subsets. For example, tumours harbouring *ALK*, *ROS1* or *RET* fusions are often characterized by an abundance of extracellular mucin, a cribriform pattern and signet-ring cell morphology[46,47].

NRG1 fusions are commonly found in invasive mucinous adenocarcinomas (IMAs), a variant of LUAD (3% of LUADs) with distinct clinical, pathological and molecular features[48]. Despite their low prevalence, IMAs comprise a sizeable proportion (28%) of all *NRG1* fusion-positive lung cancers[49,50]. *NRG1* fusion-positive IMAs tend to have higher-risk features and are associated with worse outcomes compared with *KRAS*-mutant IMAs[49]. IMAs lacking *NRG1* fusions typically harbour a wide range of *KRAS* mutations, especially G12D/V, and other driver alterations (including non-*NRG1* fusions) found in non-mucinous LUADs.

In contrast to other mitogenic drivers, *MET* exon 14 skipping is associated with several rare histological subtypes of NSCLC, namely sarcomatoid carcinomas and adenosquamous carcinomas. Most *MET* exon 14-altered tumours are LUADs, although the frequency of sarcomatoid and adenosquamous histologies can be four to six times higher in patients with *MET* exon 14-altered LUADs than in patients with *MET* wild-type disease[51,52]. These histological variants are similarly enriched among patients with highly *MET*-amplified lung cancers, suggesting a link with broader MET activation and histology predilection[51,52]. Other clinical–genotype associations include increased thrombotic risk in patients with NSCLC positive for *ALK* or *ROS1* fusions[53,54].

Many rare oncogenic alterations tend to occur predominantly in younger never-smokers or former light smokers. Racial and ethnic differences might also exist, although these are less well studied for many rare oncogenes. *EGFR* and *ERBB2* exon 20 mutations, which phenocopy classic *EGFR* mutations, are more commonly found in female never-smokers of Asian ethnicity[11]. Fusions are typically found in patients with little to no cigarette smoking history[55–57]. By contrast, *MET* exon 14 alterations are commonly diagnosed in older patients (median age 72 years) with more substantial smoking histories (>10 pack-years), including those who have smoked heavily (>20 pack-years)[58]. Transversion mutations involving *KRAS* (such as G12A and G13C) and *MAP2K1* (ref. [41]) (such as K57N) might be enriched in former and current smokers[59].

Whether some of the observations related to smoking history are sociologically conditioned is currently unclear. For example, older patients might have commenced smoking during their youth when smoking was endemic. Younger patients might never have smoked owing to clearer public health messaging, particularly over the past few decades.

Activity of chemotherapy and immunotherapy. Rare molecular subtypes of NSCLC can be broadly divided into two groups on the basis of responsiveness to chemotherapy. In the first group, chemotherapy can provide durable benefit. According to data from retrospective studies, regimens containing pemetrexed can result in higher objective response rates (ORRs) and longer median progression-free survival (PFS) in patients with *ALK*, *ROS1* or *RET* fusion-positive cancers compared with those in patients with other alterations, such as *KRAS* or *EGFR* mutations[60–62]. In the second group, retrospective data suggest more modest levels of benefit. This group includes *BRAF*[63], *ERBB2* (ref. [64]) and *EGFR* exon 20 mutations[65], and *NTRK1/2/3* (ref. [66]) and *NRG1* (ref. [67]) fusions.

The majority of patients with oncogene-driven NSCLC do not respond to immune-checkpoint inhibitors (ICIs) administered as single agents (ORRs 0% in patients with *ALK* fusions, increasing to 26% in those with *KRAS* mutations), and median PFS is short in these patients[68] (2.1 months in patients with *RET* fusions, increasing to 3.4 months in those with *MET* exon 14 alterations). This poor activity is possibly owing to a poorly immunogenic microenvironment, a lower tumour mutational burden (TMB) than that of driver-negative cancers, limited $CD8^+$ T cell infiltration and/or other factors[69].

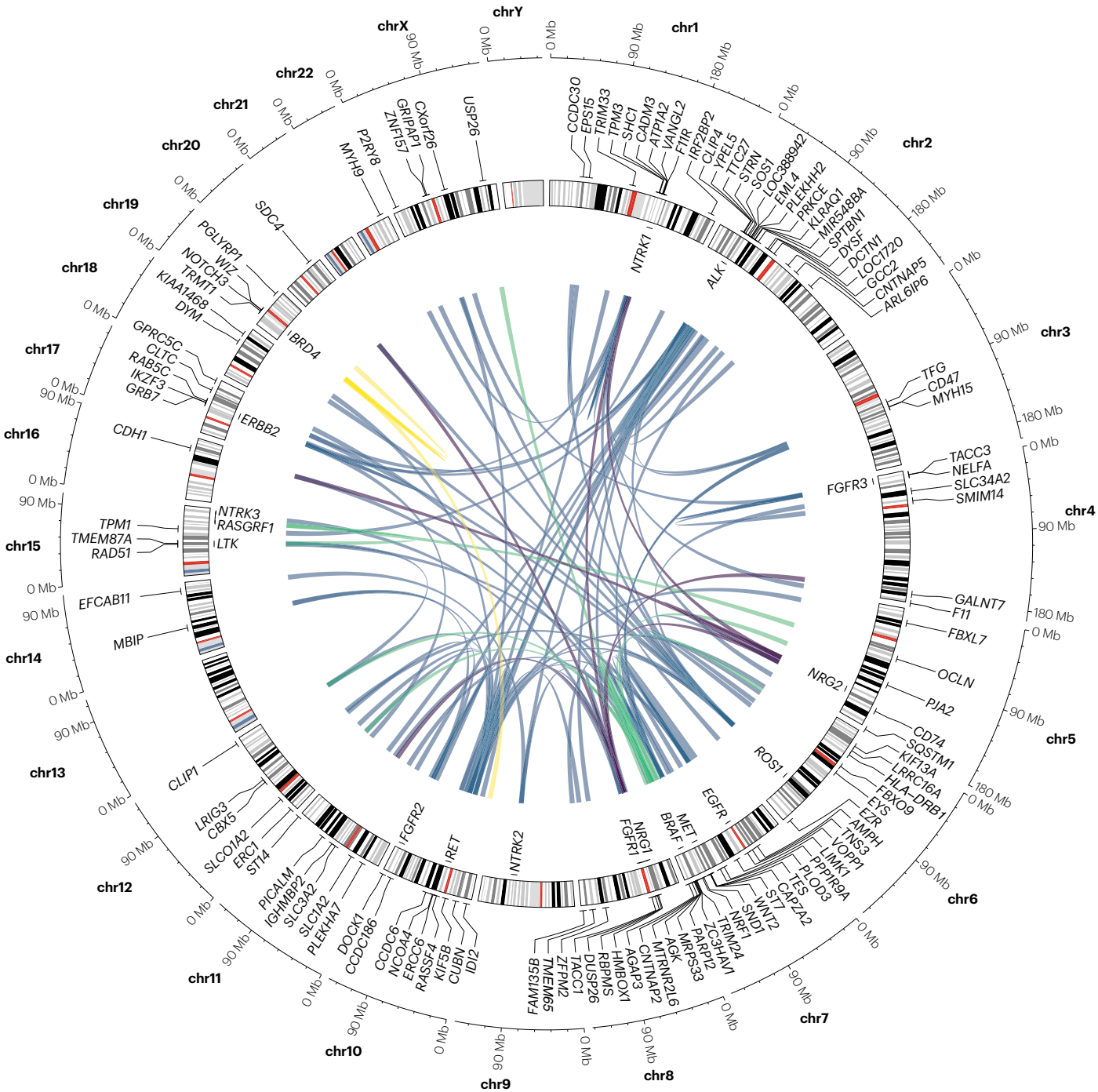

Fig. 4 | Fusion circos plot. Intrachromosomal and interchromosomal fusions involving receptor tyrosine kinase genes (blue), MAPK pathway members *RASGRF1* and *BRAF* (green), *NRG1/2* and *BRD4* (yellow) genes are shown. These genes are shown in the inner track, and the fusion partners are shown in the outer track of the circos plot. Common fusions in lung cancer include *EML4-ALK*, *KIF5B-RET* and *CD74-ROS1*; partner preference might be influenced by intrinsic genome stability, susceptible loci and transcriptional activation. The fusions have been curated from publicly available databases with RNA sequencing including the The Cancer Genome Atlas cohort[193] and MSK-IMPACT and MSK-Fusion cohort from GENIE (v12)[13].

Nonetheless, improved levels of clinical benefit and even long-term responses have been observed in certain scenarios. Smokers with *BRAF*-mutant lung cancers have a longer median PFS duration than never-smokers with lung cancers harbouring alterations in the same gene (4.1 versus 1.9 months[68]). Another example is provided by the finding that patients with metastatic *MET* exon 14-altered, PD-L1-high

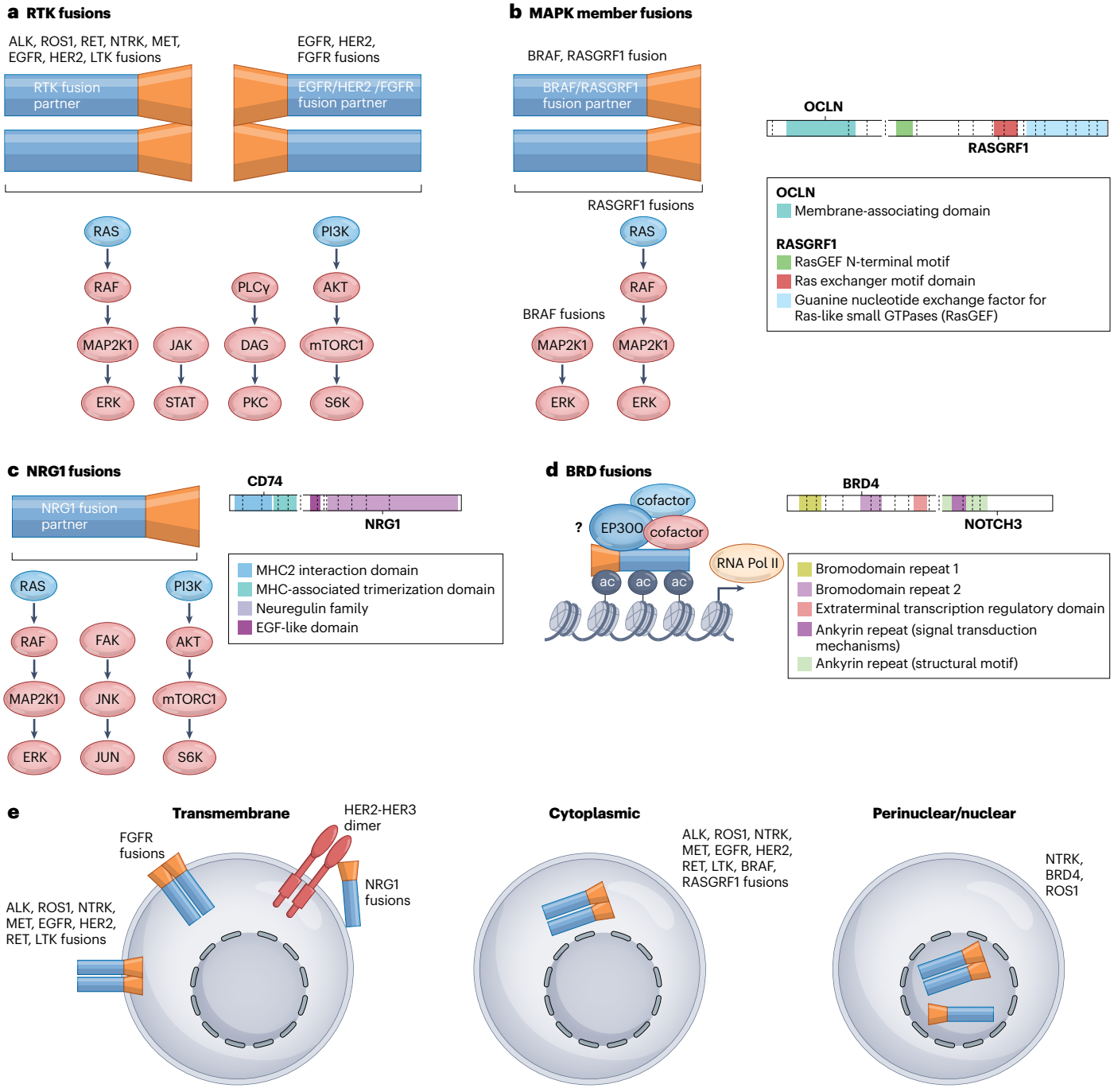

Fig. 5 | Fusions structure, signalling and localization. a, Fusions involving receptor tyrosine kinase (RTK) genes are shown. Classic 3′ fusions harbour the RTK in the downstream position. These include *ALK*, *RET*, *ROS1*, *NTRK1/2/3* (more commonly *NTRK1/3* than *NTRK2*), *ERBB1/2/4*, *MET* and *LTK* (such as *CLIP1-LTK*) fusions. 5′ fusions harbour the RTK upstream of the fusion partner, including those involving the RTKs *FGFR1/2/3*. Fusions can lead to activation of MAPK, JAK–STAT, PLCγ and PI3K–AKT pathways. **b**, Fusions involving MAPK pathway members (*RASGRF1* and *BRAF*) are shown. These putatively signal through MEK–ERK. Many *RASGRF1* fusions (such as *OCLN-RASGRF1*) feature a transmembrane domain that anchors the RAS–GEF domain to the cell membrane, facilitating RAS activation. *BRAF* fusions (such as *TRIM24-BRAF* and *LIMD1-BRAF*) often include the BRAF kinase in the 3′ position. **c**, *NRG1* and *NRG2* fusions can additionally signal through FAK–JNK–JUN. NRG1 (commonly CD74–NRG1 and SLC3A2–NRG1) and NRG2 (CD74–NRG2α) serve as ligands for ERBB family members; NRG2 fusions may preferentially activate ERBB4. **d**, BRD4–NOTCH3 includes the bromo and extraterminal domains of BRD4, which sequester histone acetyltransferases and other transcriptional co-factors to chromatin regions that transcribe pro-proliferative and anti-differentiation genes, and the active ANKYR domain of NOTCH3. The contribution of each of these domains to the oncogenic activity of the BRD4–NOTCH3 fusion is unknown. **e**, The cellular localization of specific fusions is shown. Note that localization may vary within fusion subsets (such as between *ROS1* fusions that can be transmembrane, cytoplasmic or perinuclear/nuclear) depending on the 5′ partner.

(tumour proportion score ≥ 50%) NSCLC have an ORR of 43% and a median duration of response of 13.9 months on first-line pembrolizumab, although the median PFS duration in these patients was 3.5 months[70]. Of note, the efficacy of chemotherapy and immunotherapy in rare molecular subsets of lung cancer is largely based on data from retrospective and small series, representing an important limitation to these analyses.

Concurrent or sequential use of ICIs and TKIs can increase toxicity compared with the use of only one of these modalities. For example, severe immune-related adverse events such as pneumonitis were observed in 15% of patients with *EGFR*-mutant NSCLC receiving concurrent osimertinib plus an anti-PD-1/PD-L1 antibody[71]. TKI therapy after an ICI can result in an increased incidence of transaminitis (grade 3–4 events in 45.5% of patients receiving crizotinib for *ALK* fusions[72]) and hypersensitivity (any-grade events in 11.2% of patients receiving selpercatinib for *RET* fusions[73]). As such, should molecular testing results not yet be available, chemotherapy can reasonably be considered over chemotherapy plus an ICI or ICI monotherapy in patients with suspected oncogene-driven lung cancers.

Diagnostics

Molecular profiling evolution

The discovery of rare oncogenic driver alterations has substantially transformed molecular testing practices in lung cancer over the past decade[74]. Genotyping approaches previously focused on a few genes with sequential testing using single-gene assays (such as PCR, fluorescence in situ hybridization (FISH) or Sanger sequencing)[75]. The most commonly altered genes (such as *KRAS* and *EGFR*) were analysed first, followed by less commonly altered genes; serial testing was performed until a positive result was found. With improvements in technology and the ever-growing list of actionable targets, diagnostic paradigms have converged on next-generation sequencing (NGS) as a more comprehensive, economical and tissue-efficient approach[76]. In turn, wider adoption of more comprehensive NGS panels that enable concurrent sequencing of a larger number of genes has both increased the likelihood of detecting rare alterations and uncovered previously undetected alterations.

Many NGS assays are designed to concurrently interrogate hundreds of genes, including rare potential drivers that were previously not prioritized owing to their limited incidence (such as *NTRK1/2/3* fusions) or investigational status (such as *NRG1* or *FGFR1/2/3* fusions)[77]. Standalone assays for rare driver alterations are not widely available and are often difficult to implement with limited tissue availability; therefore, NGS often provides the only screening method for these variants. Within commonly tested genes (such as *EGFR*), NGS can distinguish uncommon genotypes (such as exon 20 insertions, which can be missed by some hotspot PCR-based assays[78]). NGS also enables the identification of multiple classes of alterations (including amplifications, mutations and fusions[33,79]) and potentially novel alterations, thus fuelling current and future research efforts.

Optimizing driver identification

RNA-based testing. Targeted DNA-based NGS is typically the primary and/or only assay used for genotyping; nonetheless, the sensitivity of this method for fusions and alternatively spliced transcripts can be variable depending on assay design, gene coverage and target enrichment[80,81]. Sequencing of introns (where most breakpoints occur) can be challenging owing to size constraints (the sheer size of *NRG1* introns precludes adequate coverage[48,67,82]) and repetitive sequences (*ROS1* intron 31 is difficult to capture owing to the presence of repetitive long interspersed nuclear elements)[80].

By contrast, RNA-based methods enable direct assessments of oncogenic RNA transcripts that lack large intronic sequences, enabling more efficient and sensitive analyses. RNA-based NGS can also enable the detection of occult kinase fusions missed by DNA sequencing[80,81], thus improving sensitivity. Furthermore, RNA-based testing optimizes specificity by confirming that certain fusions of unknown significance detected in DNA are not transcribed into oncogenic fusions, while also confirming that others produce novel chimeric transcripts[83].

For splice site alterations, DNA hybrid capture-based target enrichment outperforms amplicon-based methods; however, the intrinsic limitations of DNA sequencing remain[33,84]. Without adequate coverage of *MET* introns flanking exon 14, large deletions and cryptic splice site mutations located deep within introns (such as those located more than ten nucleotides from intronic–exonic junctions) can be missed[85]. Furthermore, DNA-based NGS occasionally reveals deep intronic variants in *MET* introns 13 and 14 that have an unclear effect on splicing. By contrast, RNA sequencing can determine which variants lead to exon 14 skipping by directly capturing aberrant splicing byproducts[86].

Despite the advantages of RNA-based sequencing for selected alterations, the unique challenges associated with using RNA as a clinical analyte must also be recognized. Compared with DNA, RNA is considerably more labile and prone to degradation, leading to clinical testing failure rates of 10–30%[87,88]. Pre-analytical strategies for optimizing RNA testing include prioritizing specimens with abundant tumour content and avoiding the use of older, archived samples that are likely to yield RNA of lower quantity and quality.

A consensus approach to integrating DNA-based and RNA-based sequencing workflows has yet to be established. While performing upfront dual DNA-based and RNA-based NGS is one strategy[89], this might not be necessary in all patients and might be prohibitive in resource-limited settings. An alternative strategy (Supplementary Fig. 2) uses DNA-based NGS as a primary screening assay with subsequent RNA-based NGS in selected patients (such as patients who are driver-negative on DNA testing, and/or those with fusions or intronic mutations of unknown significance[81]). This model focuses on addressing the limitations of DNA-based testing and might facilitate the more judicious and cost-effective use of RNA-based testing, albeit with longer total turnaround times for scenarios requiring sequential testing.

Copy-number analysis. In addition to fusions and splice alterations, rare gene amplifications can be difficult to capture using DNA-based NGS owing to a variety of biological, pre-analytical and post-analytical factors. Importantly, the detection of amplifications relies heavily on the degree of amplification, tumour purity and clonality of the sample. For example, if tumour cells harbouring high-level *MET* amplifications are admixed with an abundance of immune cells and stroma, the predominance of non-tumour cells can obscure the *MET* amplifications.

Lower levels of amplification and/or the presence of amplifications in only a subset of tumour cells can further decrease the level of sensitivity. Data from dilutional studies indicate that detection of amplifications might not be reliable in specimens with <20% tumour content[90]. Hence, low-purity tumour samples with negative or equivocal amplification results might benefit from further testing using FISH, which can provide a more granular analysis at single-cell resolution.

The interpretation of copy number results from NGS is further complicated by the lack of established thresholds for amplification and guidelines for the selection of therapy. For example, the presence

of high-level *MET* amplifications enriches for responses to MET inhibition[91,92], although the definition of such amplifications can vary substantially between assays, and clinical activity can be seen across various copy states[90]. Further standardization is needed to enable the implementation of therapeutically relevant cut-offs for rare actionable amplifications.

Liquid biopsies. Adequate tumour tissue samples are essential for successful NGS; however, samples acquired via invasive procedures are not always sufficient for comprehensive testing[93]. Liquid biopsies using approximately 3–10 ml of plasma and the subsequent analysis of circulating tumour DNA (ctDNA) involve shorter analysis turnaround times and can serve as a useful alternative to tissue-based genotyping when a tumour biopsy is not safe or feasible[94]. Despite the well-recognized utility of ctDNA testing, several important issues must be considered.

Given the scarcity of plasma ctDNA and the need for ultra-deep sequencing (usually >10,000× depth), liquid biopsy panels typically include fewer genes than tissue-based panels in order to balance sequencing breadth and depth[95]. As a result, genes with highly recurrent alterations are often prioritized, and less commonly altered genes are sometimes excluded. An example is provided by the MSK-ACCESS liquid biopsy panel, which covers 129 genes. By contrast, its tissue-based counterpart MSK-IMPACT includes 505 genes in its current iteration. Other commercial ctDNA-based assays such as Guardant360 have a highly focused panel of <100 genes[94] while Tempus xF and FoundationOne Liquid, despite their larger gene panels, still cover fewer genes than their tissue-based NGS counterparts[96,97].

Compared with plasma, other bodily fluids (such as cerebrospinal fluid or pleural effusions) can be enriched in ctDNA, enabling analysis using less-sensitive NGS assays with the larger and more inclusive panels that are generally reserved for tissue-based sequencing[98,99]. Nonetheless, ctDNA testing has variable levels of sensitivity regardless of the source, and all negative results should be confirmed by tumour tissue testing[94].

The limitations of DNA-based NGS analysis of tissue samples also apply to ctDNA-based testing. Currently, no clinical assays for analysis of RNA-based liquid biopsies are available for use in patients with lung cancer (tissue is currently required for RNA-based detection of fusions and exon skipping), although notable advances in circulating tumour cell, cell-free and exosomal[100–103] RNA profiling suggest that such assays might be incorporated into clinical workflows in the future[100–103].

Novel driver discovery

Targeted DNA-based and RNA-based NGS sometimes fail to identify a clear oncogenic driver within the RTK–RAS–RAF pathway. While a distinct driver-negative subset characterized by smoking-induced malignancies with complex genomics, a high TMB, and/or alterations in *TP53*, *STK11* or *KEAP1* alterations exists[44], the absence of a driver on targeted NGS, particularly in never-smokers with a low TMB, could be a false-negative result that justifies more comprehensive analysis using whole-transcriptome sequencing (WTS) or whole-genome sequencing (WGS).

WTS of supposedly 'driver-negative' tumours has enabled the discovery of fusions containing *NRG2* (refs. [19,49]), *RASGRF1* (ref. [16]) and *LTK*[104] and clinical-grade WTS might facilitate a more unbiased search for other rare or novel fusions compared with targeted panel-based profiling. Similarly, WGS might also enable novel oncogenic signatures to be uncovered. In the TCGA LUAD project, WGS of tumours deemed driver-negative on WTS and whole-exome sequencing revealed the presence of pathogenic copy number alterations, complex rearrangements and non-coding alterations including a candidate driver mutation in the *ILF2* promoter region[105]. Furthermore, WGS enabled the detection of canonical drivers missed by whole-exome sequencing owing to low tumour purity and/or poor coverage, thus highlighting the importance of pre-analytical factors and quality control metrics[105].

While WTS and WGS are not routinely used in the clinic, data from studies using this method suggest an emerging role in biomarker discovery in driver-negative tumours (Supplementary Fig. 2), especially those deemed more likely to harbour occult drivers (such as those with a low TMB that are also not smoking-related). The potential utility of other multi-omics approaches (such as methylomics and proteomics) continues to evolve.

Targeted therapy

Drug classes

Small molecules. Kinase inhibitors comprise most of this group. Kinase substrates can be classified into tyrosine kinases (EGFR, HER2, MET, ALK, RET and ROS1), serine–threonine kinases (BRAF) and dual-specificity kinases (MAP2K1 and MAP2K2). TKIs (such as mobocertinib, capmatinib, tepotinib, alectinib, brigatinib, lorlatinib, selpercatinib, pralsetinib, crizotinib and entrectinib) are the most common class of approved agents (Tables 1,2) for oncogene-driven lung cancers. The remaining minority of approved or guidelines listed agents include serine–threonine kinase inhibitors (dabrafenib and vemurafenib) and dual-specificity kinase inhibitors (trametinib)[106].

Kinase inhibitors can be classified by mechanism of action. Interestingly, all FDA-approved TKIs are ATP-competitive type I inhibitors that target kinases in their active conformation. ATP-competitive type II inhibitors (such as cabozantinib) target kinases in their inactive conformation and are less common; none are approved for oncogene-driven lung cancers. Type III inhibitors (such as trametinib) are non-ATP competitive allosteric inhibitors[107]. Generations have also been assigned to kinase inhibitors that target a single molecular subset of lung cancers (such as *ALK* fusion-positive NSCLC). Later-generation agents often have additional features relative to their earlier-generation counterparts, such as improved central nervous system (CNS) activity and activity against resistance mutations.

Several novel small molecules have either entered or are expected to enter clinical trials. Dabrafenib and vemurafenib target monomeric BRAF V600E-mutant forms of BRAF, whereas newer RAF inhibitors that target dimers (such as PLX8394 (ref. [108]) and BGB-3245 (NCT04249843) are being investigated in patients with non-V600E (for example class II) *BRAF*-mutant malignancies. Agents designed to deliver targeted protein degradation (such as proteolysis-targeting chimeras (PROTACs) and molecular glues) are also currently being explored in certain oncogene-driven lung cancers such as those harbouring *KRAS*, *EGFR*, *MET* or *BRAF*[109] mutations, or *ALK* or *RET* fusions[110]. Although effective small molecules against these alterations have been developed and approved, PROTACs are mechanistically different and may address complex on-target resistance by degrading proteins that are unlikely to bind approved or investigational small molecules (such as TKIs) owing to resistance mutations that confer steric hindrance[111]. In addition, PROTACs may be active against alterations for which there are no currently approved drugs, such as non-G12C *KRAS* mutations.

Antibody-based therapies. Apart from small molecules, antibody-based therapies are another widely used class of agents. Unconjugated antibodies can be monospecific or bispecific. Monospecific antibodies harbour specificity for a single antigen or epitope (such as trastuzumab

Table 1 | Clinical activity of targeted therapies against 'rare' mutations and copy number alterations in lung cancers

Molecular alteration	Agent	Patient population	Mechanism of action	ORR (%)	Median DoR (months)	Median PFS (months)	Median OS (months)
EGFR exon 20 insertions	Amivantamab[a,b] (ref. [120])	Pretreated	EGFR–MET BiAb	40	11.1	8.3	22.8
	Mobocertinib[a,b] (ref. [119])	Pretreated	EGFR TKI	28	17.5	7.3	24
	Poziotinib[186]	Treatment-naive and pretreated	EGFR and HER2 TKI	32	8.6	5.5	19.2
	CLN-081[c] (ref. [157])	Pretreated	EGFR TKI	38	10	10	Not mature
BRAF V600E mutations	Dabrafenib + trametinib[a,b] (ref. [118])	Treatment-naive	BRAF S/TKI and MAP2K1/2 inhibitor	64	10.4	10.9	24.6
	Dabrafenib[b] (ref. [126])	Treatment-naive and pretreated	BRAF S/TKI	33	9.9	5.5	12.7
	Vemurafenib[b] (ref. [127])	Treatment-naive and pretreated	BRAF S/TKI	37	7.2	6.5	15.4
ERBB2 mutations	Trastuzumab deruxtecan[a,b] (ref. [115])	Pretreated	HER2 ADC	55	9.3	8.2	17.8
	Trastuzumab emtansine[b] (ref. [124])	Treatment-naive and pretreated	HER2 ADC	44	4	5	NR
	Trastuzumab + pertuzumab + docetaxel[187]	Pretreated	HER2 mAb and chemotherapy	29	11	6.8	17.6
	Poziotinib[c,123]	Treatment-naive and pretreated	EGFR and HER2 TKI	27	5	5.5	15
	Pyrotinib[122]	Pretreated	Pan-HER TKI	30	6.9	6.9	14.4
ERBB2 copy number increases	Pyrotinib[152]	Treatment-naive and pretreated	Pan-HER TKI	22	7.2	6.3	12.5
	Pertuzumab + trastuzumab[151,188]	Pretreated (included *ERBB2* amplification/overexpression)	HER2 mAb	13	NR	NR	NR
MET exon 14 alterations	Capmatinib[a,b] (ref. [92])	Treatment-naive	Type Ib MET TKI	68	12.6	12.4	NR
		Pretreated	Type Ib MET TKI	41	9.7	5.4	NR
	Tepotinib[a,b] (ref. [117])	Treatment-naive and pretreated	Type Ib MET TKI	46	11.1	8.5	17.1
	Crizotinib[b] (refs. [149,158])	Treatment-naive and pretreated	Type Ia MET TKI	32	9.1	7.3	20.5
	Savolitinib[189]	Treatment-naive and pretreated	Type Ib MET TKI	47	NR	6.8	12.5
	Amivantamab[121]	Treatment-naive and pretreated	EGFR–MET BiAb	33	NR	NR	NR
MET copy number increases	Capmatinib[b] (ref. [92])	Treatment-naive (GCN ≥ 10)	Type Ib MET TKI	40	7.5	4.2	NR
		Pretreated (GCN ≥ 10)	Type Ib MET TKI	29	8.3	4.1	NR
	Tepotinib[b] (ref. [150])	Treatment-naive and pretreated (*MET* copy number ≥ 2.5)	Type Ib MET TKI	42	NR	4.2	NR
	Crizotinib[b] (ref. [149])	Treatment-naive and pretreated (MET/CEP7 ≥ 4)	Type Ia MET TKI	38	5.2	6.7	11.4

ADC, antibody–drug conjugate; BiAb, bispecific antibody; DoR, duration of response; GCN, gene copy number; mAb, monoclonal antibody; PFS, progression-free survival; NR, not reported; ORR, objective response rate; OS, overall survival; S/TKI, serine–threonine kinase inhibitor; TKI, tyrosine kinase inhibitor. [a]FDA-approved. [b]Included in the National Comprehensive Cancer guidelines. [c]Fast track/breakthrough designation by FDA.

for HER2 (ref. [112]) or seribantumab for HER3). Bispecific antibodies target two antigens or epitopes (for example, amivantamab for MET and EGFR or zenocutuzumab for HER3 and HER2). These antibodies have a variety of functions including interference with ligand binding, inhibition of RTKs and induction of antibody-dependent cytotoxicity.

Expanding the scope of antibody-based targeting, antibody–drug conjugates (ADCs) have emerged as a new class of agents. ADCs consist of an antibody (typically a class 1 IgG), a cytotoxic payload (such as an auristatin, maytansinoid, calicheamicin or camptothecin), and a linker (cleavable or non-cleavable) that connects the two components[113]. New payloads with putative immunomodulatory effects (such as TLR7/8 agonists[114], known as immune-stimulating antibody conjugates) have entered clinical testing. ADCs explored in patients with oncogene-driven lung cancers include the anti-HER2 ADCs trastuzumab emtansine and trastuzumab deruxtecan[115], and the anti-MET ADC telisotuzumab vedotin[116].

Activity

The therapeutic actionability of rare molecular subtypes of lung cancer varies by alteration. At one end of the spectrum lie alterations for which targeted therapies are approved in one or more countries. Even within this group, variations in response rate (indicating the degree of actionability) and level of evidence supporting approval (phase I/II versus phase III trials) are observed. At the other end of the spectrum lie alterations for which clinical actionability remains questionable. Including discussions of all possible therapies for each alteration is currently not feasible, therefore a summary of data from preclinical and clinical studies is provided (Tables 1,2, Supplementary Table 4).

Mutations. The only rare-oncogene-driven lung cancers for which targeted therapies are approved and/or listed in clinical guidelines are *EGFR* exon 20-mutant, *ERBB2*-mutant, *BRAF* V600E-mutant and *MET* exon 14-altered NSCLC. These various classes of drugs are clinically

active and have provided benefit in many patients; however, none of these has consistently and simultaneously resulted in an ORR of >50% and a median PFS of >1 year either as monotherapy or combination therapy. In treatment-naive patients with *MET* exon 14 alterations, capmatinib results in an ORR of 68% and a median PFS of 12.4 months[92], although other members of the same drug class have more limited activity (tepotinib, which results in an ORR of 46% and a median PFS of 8.5 months[117], or the broad-spectrum TKI crizotinib, which results in an ORR of 32% and a median PFS of 7.3 months).

Trastuzumab deruxtecan for *ERBB2*-mutant lung cancers (ORR 55%, median PFS 8.2 months)[115] and dabrafenib plus trametinib for *BRAF* V600E-mutant lung cancers (ORR 64%, median PFS 10.9 months[118]) are among the more active approved drugs for lung cancers harbouring rare mutations. The rest of the therapy–oncogene pairs are typically associated with more modest ORRs of 30–50% and median PFS durations of <1 year: mobocertinib[119] or amivantamab[120] in patients with *EGFR* exon 20 mutations, and capmatinib (in previously treated patients)[92] or tepotinib[117] in patients with *MET* exon 14 alterations.

In a field previously dominated by small molecules, proof of principle that antibody-based therapies can have comparable or even improved levels of activity is growing. An example is provided in patients with *EGFR* exon 20 mutations, for whom both the antibody amivantamab and the small molecule mobocertinib are available, with similar levels of activity observed in separate studies. Amivantamab has also demonstrated activity (ORR 64% in a cohort including patients who had previously received a MET TKI) in patients with *MET* exon 14-altered NSCLC[121]. *ERBB2*-mutant NSCLC provides an excellent example of an improvement in activity when moving from TKIs (ORRs 0–30%[122,123]) to ADCs (trastuzumab emtansine (ORR 44%) and trastuzumab deruxtecan (ORR 55%)[115,124]).

Other mutation-driven lung cancers might be targetable based on preclinical data or case reports. For example, patients with *DDR2*-mutant tumours, which are thought to require SRC, have responded to the SRC inhibitor dasatinib[12]. Following the success of direct KRAS G12C inhibitors, other mutation-specific or pan-RAS inhibitors are emerging for the treatment of tumours harbouring non-G12C *KRAS* mutations[125].

Beyond single-agent therapies, combination therapies might be effective against tumours harbouring other RAS–MAPK pathway alterations (as observed with BRAF plus MEK inhibitors compared with BRAF inhibitor monotherapy[126,127] in patients with *BRAF* V600E-mutant cancers). Similarly, data from preclinical models of *RIT1*-mutant cancer[128] indicate a response to MEK and PI3K inhibition. Acknowledging their RAS dependence, patients with advanced-stage solid tumours

Table 2 | Clinical activity of targeted therapies against 'rare' fusions in lung cancers

Molecular alteration	Agent	Patient population	Mechanism of action	ORR (%)	Median DoR	Median PFS (months)	Median OS (months)
ALK fusions	Crizotinib[a,b] (ref. [136])	Treatment-naive	1st generation ALK TKI	74	11.3	10.9	Not reached
	Ceritinib[a,b] (ref. [137])	Treatment-naive	2nd generation ALK TKI	73	23.9	16.6	Not reached
	Alectinib[a,b] (refs. [139,138])	Treatment-naive	2nd generation ALK TKI	83	28.1	34.8	Not reached
	Brigatinib[a,b] (ref. [140])	Treatment-naive	2nd generation ALK TKI	71	33.2	24	Not reached
	Ensartinib[190]	Treatment-naive	2nd generation ALK TKI	74	Not reached	25.8	Not reached
	Lorlatinib[a,b] (ref. [141])	Treatment-naive	3rd generation ALK TKI	76	Not reached	Not reached	Not reached
RET fusions	Selpercatinib[a,b] (ref. [134])	Treatment-naive	RET TKI	85	Not reached	Not reached	Not reached
		Pretreated	RET TKI	64	17.5	16.5	Not reached
	Pralsetinib[a,b] (ref. [135])	Treatment-naive	RET TKI	70	9	9.1	Not reached
		Pretreated	RET TKI	61	Not reached	17.1	Not reached
ROS1 fusions	Crizotinib[a,b] (ref. [132])	Treatment-naive and pretreated	ROS1 TKI	72	24.7	19.3	51.4
	Ceritinib[b] (ref. [191])	Pretreated	ROS1 TKI	62	21	9.3	24
	Entrectinib[a,b] (ref. [133])	Treatment-naive	ROS1 TKI	68	20.5	15.7	47.8
	Lorlatinib[b] (ref. [160])	Treatment-naive and pretreated	ROS1 TKI	62	25.3	21	Not reached
	Repotrectinib[a] (ref. [192])	Treatment-naive	ROS1 TKI	79	NR	NR	NR
NTRK1/2/3 fusions	Larotrectinib[a,b] (ref. [129])	Treatment-naive and pretreated	TRK TKI	83	Not reached	Not reached	40.7
	Entrectinib[a,b] (ref. [156])	Treatment-naive and pretreated	TRK TKI	64	19.9	14.9	Not reached
NRG1 fusions	Zenocutuzumab[c] (ref. [143])	Treatment-naive and pretreated	HER2–HER3 BiAb	35	9.1 (pan-tumour; NR in the NSCLC cohort)	NR	NR
	Seribantumab[c] (ref. [144])	Pretreated	ERBB3 mAb	36	Not reached	NR	NR

BiAb, bispecific antibody; DoR, duration of response; mAb, monoclonal antibody; NR, not reported; NSCLC, non-small-cell lung cancer; ORR, objective response rate; OS, overall survival; PFS, progression-free survival; TKI, tyrosine kinase inhibitor. [a]FDA-approved. [b]Included in the National Comprehensive Cancer guidelines. [c]Fast track/breakthrough designation by FDA.

harbouring class III *BRAF* alterations are receiving the combination of MEK and SHP2 inhibitors in a large-cohort phase I clinical trial (NCT04800822).

Fusions. As opposed to mutation-driven lung cancers, fusion-driven lung cancers often respond well to approved targeted therapies with ORRs of >50% and median PFS durations of >1 year seen with several agents in the TKI-naive and/or treatment-naive settings (Table 2). Patients with *ALK*, *RET*, *ROS1* or *NTRK1/2/3* fusion-positive lung cancers have ORRs of 61–83%. The durability of these responses is equally impressive, with median PFS durations ranging from 9 to 35 months[129–141].

Sequential use of TKIs can be effective in patients with fusion-positive cancers. *ALK* fusion-positive NSCLC is the only subset for which this paradigm has corresponding drug approvals (for example, lorlatinib in patients who have received a previous ALK TKI, based on an ORR of 39% and a median PFS of 9.6 months)[142]; however, clinical responses to next-generation TKIs have been documented after progression on initial TKI therapy in patients with other fusion-positive lung cancers (such as in patients receiving repotrectinib for *ROS1* fusions, TPX-0046 for *RET* fusions or selitrectinib for *NTRK* fusions).

The experience with *NRG1* fusions demonstrates the utility of antibody-based therapies in fusion-positive lung cancers with putative cell-surface expression of a chimeric oncoprotein. Although these tumours depend on HER3–HER2 dimers for growth, the pan-ERBB TKI afatinib has unimpressive overall activity (median PFS 2.8 months, despite a 25% ORR)[67]. By contrast, the antibodies zenocutuzumab or seribantumab are associated with ORRs of ~35%, with a median duration of response of 9.1 months observed with zenocutuzumab in a mixed cohort of patients with *NRG1* fusion-positive solid tumours including NSCLC[143,144]. As *NRG1* fusion-positive cancers can overexpress HER3, the use of ADCs could be considered, recognizing that HER3-targeted ADCs (such as patritumab deruxtecan) have already been explored in other HER3-expressing NSCLCs, including in *EGFR*-mutant NSCLCs[145].

Patients with tumours harbouring other fusions might also benefit from targeted therapies. *MET* fusion-positive NSCLCs, many of which feature both exon 14 exclusion and an intact kinase domain can respond to crizotinib[146]. Similarly, a patient with *FGFR2* fusion-positive NSCLC responded to the pan-FGFR inhibitor erdafitinib, according to a case report[147]. Likewise, tumours with fusions involving *LTK* (whose kinase domain is 80% identical to that of ALK) can respond to ALK TKIs such as lorlatinib[104]. Finally, *RASGRF1* fusion-driven cancers can respond to MAPK pathway inhibitors (such as sunitinib) both in preclinical models and clinically[148]. *BRAF* fusions are considered class II *BRAF* alterations and patients with tumours harbouring such alterations are receiving RAF dimer or pan-RAF inhibitors in clinical trials (NCT02428712)[108].

Amplifications. Data on targeted therapies for patients with amplification-driven NSCLC are currently limited. Crizotinib, capmatinib and tepotinib are all listed in guidelines for the management of patients with lung cancers harbouring high-level *MET* amplifications[92,149,150] (Table 1). As implied by the indication, higher levels of *MET* amplification or gene copy numbers are correlated with higher response rates to TKIs. Limited activity of TKIs and antibody-based therapies has been described in patients with *ERBB2*-amplified NSCLCs[151,152]. While robust clinical evidence has yet to emerge, other RTK amplifications could be targeted using TKIs or antibody-based therapies, although the contributions of co-occurring alterations and amplifications of differing extents and focalities should be explored, as they have been for RTKs such as MET[90].

Contemporary features

Selectivity. Several rational enhancements in drug design have improved the clinical outcomes of patients with oncogene-driven lung cancers (Fig. 6). Increased target selectivity in particular is a favourable feature. In patients with *RET* fusion-positive NSCLCs, the movement from multikinase inhibitors with anti-RET activity (such as cabozantinib[153] and vandetanib[154]) to highly RET-selective agents (such as selpercatinib[134] and pralsetinib[135]) has resulted in an increase in tolerability owing to the avoidance of inhibition of non-RET kinases such as VEGFR2 (ref. [155]) and activity, in part reflecting the ability to tolerate more meaningful plasma exposures and better target coverage. Increasing ROS1 selectivity (such as with the ROS-selective inhibitor NVL-520) might avoid the adverse effects associated with TRK and ROS1 inhibitors such as entrectinib and repotrectinib, including dizziness, weight gain and withdrawal pain[156].

Increased selectivity for certain mutations is another favourable drug design feature. An example of this effect is provided by mobocertinib[119], which is more selective for EGFR exon 20-mutant proteins than other EGFR alterations. Newer agents designed to target EGFR exon 20 alterations, such as CLN-081, combine both mutant selectivity and target selectivity (inhibition of HER2 is avoided)[157].

Although largely favourable, increasing kinase selectivity might have unwanted clinical consequences with certain agents. For example, the move from using type Ia inhibitors (such as crizotinib)[158] to more potent and selective type Ib MET inhibitors (such as tepotinib) in patients with *MET*-mutant and/or amplified NSCLCs was associated with an increase in lower extremity oedema[92,117]. Similar isoform-selectivity problems might emerge with novel FGFR2- or FGFR3-selective TKIs, which may lead to cutaneous adverse effects in patients with *FGFR2*- or *FGFR3*-amplified NSCLC[147].

CNS coverage. NSCLCs have a proclivity to metastasize to the brain; the lifetime risk of brain metastases in patients with oncogene-driven lung cancers can be substantial (35–50%)[159]. Rational drug design has enabled the development of systemic therapies with improved levels of CNS coverage, particularly small molecules. TKIs for *ALK*, *RET* or *ROS1* fusion-positive lung cancers (including alectinib[138], brigatinib[140], lorlatinib[160], entrectinib[133,160], selpercatinib[134] and pralsetinib[135]) provide the best examples of such improvements, leading to intracranial ORRs of ~60–80% with activity even seen in patients with leptomeningeal disease. Randomized phase III trials have demonstrated the intracranial superiority of next-generation ALK TKIs compared with crizotinib[138,140,141].

Antibody-based therapies can also induce intracranial responses[161] although agents from this class might have impaired CNS activity owing to certain size constraints. Combining small molecules and antibodies has been investigated in patients with tumours harbouring classic *EGFR* alterations (for example, the combination of lazertinib plus amivantamab[162]) and such a paradigm could be applied to rare-oncogene-driven NSCLCs. Developing nanoparticles conjugated to cytotoxic therapies[163], effectively smaller equivalents of ADCs, might be another method for improving CNS drug delivery.

Resistance anticipation. Generational improvements in TKIs often include activity against resistance mutations that emerge with earlier-generation TKIs. For fusion-positive NSCLCs, these advances can

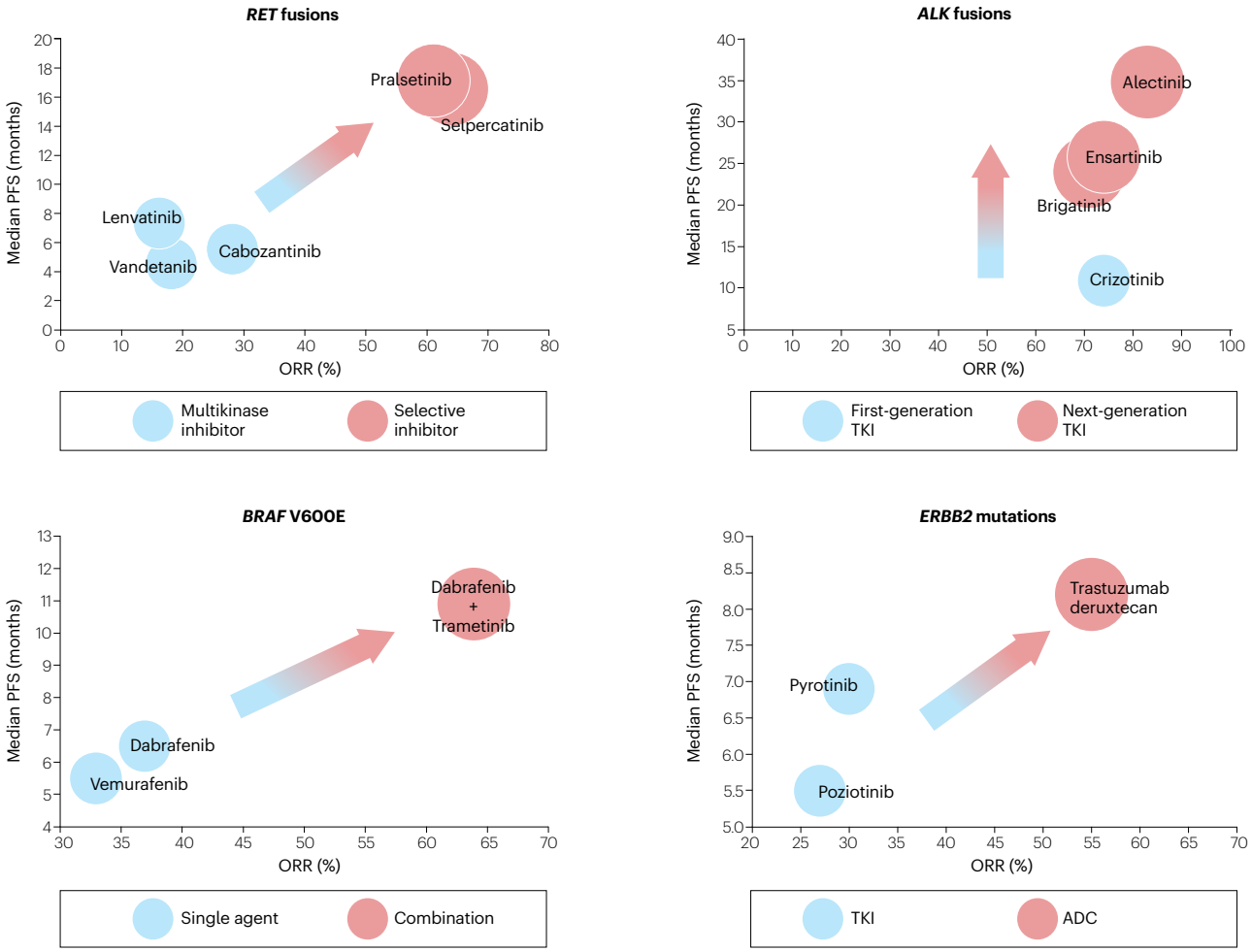

Fig. 6 | Rational drug design trends. Improvements in clinical outcomes accompanied by advances in rational drug design are depicted in these bubble plots. Objective response rates (ORR) are shown on the *x*-axes. Median progression-free survival (PFS) durations are shown on the *y*-axes. Each circle represents a specific targeted therapy strategy, including single agents and combination therapies. **a**, In *RET* fusion-positive lung cancers, both ORR and median PFS improved with the move from multikinase inhibitors[153,154,199] with anti-RET activity to the highly selective RET inhibitors, selpercatinib and pralsetinib[134,135], which entered clinical testing in 2017. **b**, Generational changes in tyrosine kinase inhibitor (TKI) use can result in substantial improvements in median PFS. Specifically, later-generation ALK TKIs (such as alectinib, brigatinib, ensartinib and lorlatinib, which is not included here owing to the median PFS not yet being reached[138,140,141,190]) with improved central nervous system activity and resistance mutation coverage have replaced the first-generation ALK TKI crizotinib based on data from randomized phase III trials[136]. **c**, The utility of combination small-molecule therapies was demonstrated by the move from single-agent BRAF inhibition with dabrafenib or vemurafenib[126,127] to the combination of a BRAF inhibitor and a MEK inhibitor (dabrafenib plus trametinib)[118]. **d**, Finally, the increase in both ORR and median PFS with trastuzumab deruxtecan[115] compared to pyrotinib and poziotinib[122,123] underscores the meaningful entry of a new wave of antibody-based targeted therapies into the clinic. ADC, antibody–drug conjugate.

include activity against gatekeeper, solvent-front and other resistance mutations. Importantly, some mutations (such as xDFG, which lead to the adoption of an inactive 'DFG out'-like state) can result in conformational resistance that precludes the activity of type I TKIs, requiring the administration of a type II TKI[164].

In patients with *ALK* fusion-positive lung cancers, improved mutational coverage is observed moving from second-generation TKIs (such as alectinib and ceritinib) to third-generation TKIs (such as lorlatinib); agents that could be considered fourth-generation TKIs that include activity against double mutations (such as TPX-0131 (NCT04849273)[165] and NVL-655 (NCT05384626)[166]) are already being tested in clinical trials. Next-generation TKIs with expanded mutation coverage are also being explored for fusions containing *RET* (TPX-0046, HM06 and LOXO-260), *ROS1* (repotrectinib, taletrectinib and NVL-520), and *NTRK1/2/3* (selitrectinib, repotrectinib and PBI-200). Notably, selected developmental programmes (such as that for repotrectinib) have followed the drug development paradigms applied to the development of ALK inhibitors and moved the clinical testing of next-generation TKIs from TKI-pretreated to TKI-naive patients[167].

Notably, rare molecular alterations might emerge as a mechanism of resistance to therapies targeting another molecular subtype. For example, oncogenic fusions (involving *ALK*, *RET*, *ROS1* or *NTRK1/2/3*)

have all been described as mechanisms of resistance to EGFR TKIs. These *EGFR*-mutant and fusion-positive cancers are a rare subset within a more common molecular subtype (*EGFR*-mutant lung cancers in general). Combination therapy regimens that involve continuing the initial targeted therapy with the addition of a second agent targeting bypass resistance can re-establish disease control in such a setting. For example, the combination of osimertinib and selpercatinib has demonstrated activity in patients with *EGFR*-mutant NSCLC that acquires *RET* fusions[168] after progression on an EGFR TKI; nonetheless, attempts to obtain regulatory approval of such a combination are confronted by similar challenges to those associated with rare single-alteration genotypes.

Research equity

A cancer population labelled as 'rare' might be associated with challenges relating to devaluation of their situation, similar to those faced by racial and/or ethnic minority groups in general society. Research into rare populations might be deemed less important to that designed to develop treatments for patients with more commonly diagnosed cancer subtypes. This issue applies to the range of frequencies observed with rare molecular subtypes of cancer (such as *NRG1* fusion-positive lung cancers, which comprise <1% of NSCLCs and might receive less attention than *ALK* fusion-positive NSCLCs that make up 3–4% of this group). Therapeutic trials might be perceived as infeasible or of lower financial value to pharmaceutical companies in this context. Fortunately, multiple stakeholders have mobilized to establish research equity for patients with less-commonly diagnosed cancer subtypes (Fig. 7).

Advocacy

The number of biomarker-specific lung cancer patient advocacy groups has risen substantially over the past decade[169] (Supplementary Table 6). This increase has occurred in parallel with the growing recognition of rare molecular subtypes of lung cancer and the development of targeted therapies for patients with these cancers. These groups include: ALK Positive (*ALK* fusions), BRAF Bombers (*BRAF* alterations), Exon 20 Group (*EGFR* and *ERBB2* exon 20 mutations), EGFR Resisters (including *EGFR* mutations beyond L858R and exon 19 deletion), KRAS Kickers (*KRAS* mutations), MET Crusaders (*MET* alterations), NTRKers (*NTRK1/2/3* fusions), RET Renegades (*RET* fusions), RET Positive (*RET* fusions) and ROS1ders (*ROS1* fusions). Initiatives have emerged such as the Biomarker Collaborative that are designed to help patients and their representatives to find the most appropriate group for a specific molecular subtype of lung cancer.

Advocates have focused on increasing the awareness of molecular subtypes of lung cancer, the availability of standard of care and investigational therapies, understanding and treatment of adverse effects and physician expertise. Research acceleration is another major goal. As an example, the Global ROS1 Initiative[170] promotes research specifically into *ROS1* fusion-positive lung cancer in several priority areas (education, basic science, real-world data, therapeutics and survivorship). Under this initiative, the ROS1 Cancer Model Project facilitates the donation of tumour specimens for the creation of patient-derived models.

Trials and regulation

Trial design. The historical approach to clinical trial design heavily favours the exploration of more common molecular subtypes of cancer within a single histology[171]. Such strategies are not appropriate for rare cancer subtypes; therefore, various master protocols have been developed to address this challenge. Umbrella trials (such as BATTLE[172] and Lung-MAP[173]) have explored matched targeted therapy cohorts for different molecular subtypes of a single histology. Several (such as the National Lung Matrix Trial[174]) include a centralized molecular screening effort. To date, many umbrella trials have been designed as signal-finding studies; none of these trials has singularly supported the approval of a targeted therapy.

Basket trial programmes, in which patients are accrued according to their molecular alteration regardless of cancer type[175], have supported regulatory approval. The seminal tumour-agnostic approvals of TRK inhibitors demonstrate how basket trials can address many of the challenges relating to drug development for low-frequency alterations[176]. Aggregating *NTRK1/2/3* fusion-positive NSCLCs with other cancers established regulatory-grade data that has enabled the approval of TRK inhibitors in at least 40 countries[177]. Nonetheless, other design features (such as seamless clinical trials[178] and the use of contemporary statistical analysis methods in adaptive trial designs[179]) have similarly hastened drug development (Fig. 7).

Regulatory support. Various health-care agencies have developed pathways (Supplementary Tables 7,8) to support drug development for rare cancers. In the USA, investigational agents might be designated as orphan drugs if developed for a population with a total prevalence of <200,000 people[1]. This designation can determine research grant eligibility, and the availability of trial tax credits and fee waivers. In Europe, drugs can obtain orphan medicinal product status if the treated condition is found in <50 out of 100,000 people. The FDA has approved more cancer drugs for orphan indications than the EMA[180].

The FDA has several programmes[181] that are designed to expedite the drug approval process: fast track, breakthrough therapy, priority review and accelerated approval[182]. Drugs can receive fast-track designation (grants increased FDA interactions and rolling review) if they are intended to treat a serious condition and address an unmet need, such as cancers with few or no effective therapeutic options. In 2010, crizotinib was the first drug to receive fast-track status for a rare-oncogene-driven NSCLC (Supplementary Table 7). Breakthrough therapy designation (established in 2012 with the added benefit of assigning an FDA review committee) requires clinical evidence demonstrating substantially improved activity relative to existing treatments. In 2013, the ALK inhibitor alectinib was the first drug to receive breakthrough therapy designation for a rare-oncogene-driven lung cancer.

Priority review, requested at the time of submission of a drug approval application, shortens the review time, the time from submission of the application to a decision being made, to 6 months or less. Accelerated approval is a conditional approval that permits the use of surrogate end points for survival such as ORR; post-approval data must then provide confirmatory evidence of benefit, after which a drug might then receive full approval. In *ALK* fusion-positive NSCLCs, both second-generation and third-generation TKIs first received accelerated approval for ALK TKI-pretreated cancers, followed by a full approval that included use in TKI-naive cancers. Trastuzumab deruxtecan (for *ERBB2*-mutant NSCLC) was the first ADC to receive accelerated approval for a rare-oncogene-driven NSCLC subtype.

Real-world evidence. Real-world data can come from various sources[183] (examples include wearables, electronic health records, reimbursement claims and billing activities) and these can be analysed to produce real-world evidence (RWE). RWE is prospectively or retrospectively curated clinical evidence regarding the use, benefits

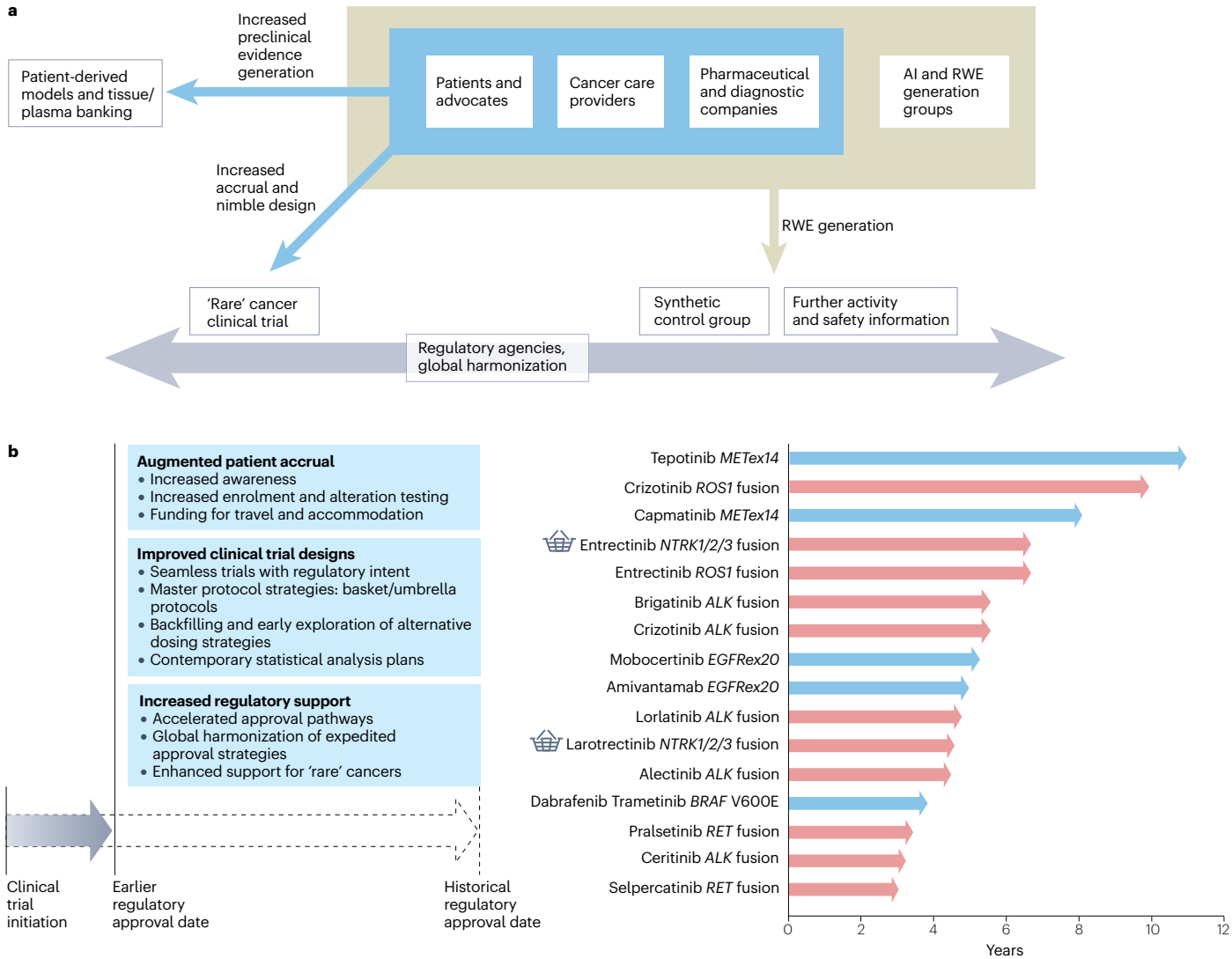

Fig. 7 | Stakeholder cooperation. a, In the field of rare cancer research, multiple stakeholders have come together to generate an increasing amount of data. These stakeholders include patients and their advocates, cancer care providers, pharmaceutical and diagnostic companies, and groups with an interest in artificial intelligence (AI) and real-world evidence (RWE). Efforts to improve model development, tissue and/or plasma sampling, trial accrual, and ultimately promote timely global targeted therapy approvals are critical. **b**, Left panel: factors that are poised to increase the speed with which molecularly matched therapeutics are approved are shown above. Right panel: time to the approval of various targeted therapies in oncogene-driven lung cancers is shown relative to the date that the first phase I trial of that agent was launched. The pink and blue arrows represent fusion- and mutation-targeted therapies, respectively. The basket symbol indicates targeted therapies that have been explored using basket trials.

(response and durability) and risks (adverse effects) associated with a particular medical product.

Stakeholders involved in the development of drugs for rare cancers have placed a premium on RWE generation. Academic investigators have formed global registries for rare-oncogene-driven NSCLCs (such as GLORY[184] for *RET* fusions and eNRGy1 (ref. [67]) for *NRG1* fusions). Patient-powered research networks leverage social media, websites and/or applications to collate patient-reported outcomes. Commercial stakeholders have aggregated large, anonymized RWE datasets.

Health-care agencies have signalled an increase in the adoption of RWE to support regulatory decision making[185], owing to movements such as the 21st Century Cures Act. In rare-oncogene-driven NSCLCs for which initiating a randomized trial is challenging, data from single-arm targeted therapy trials might be compared to synthetic standard-of-care cohorts in a molecularly enriched population. The digitalization of structured health-care data, natural language processing and artificial intelligence are all likely to accelerate these efforts.

Conclusions

Lung cancer remains an archetypal example of a tumour type that is enriched for rare oncogenes. These molecular subtypes of lung cancer have challenged our conceptions of mechanisms of oncogenesis and

reshaped our approach to molecular diagnostics. Importantly, multiple stakeholders have responded to the increasing clinical identification of these rare alterations by placing a premium on advocacy, expanded data generation, rational drug discovery and global regulatory openness to expediting therapeutic approvals.

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References

1. Tambuyzer, E. et al. Therapies for rare diseases: therapeutic modalities, progress and challenges ahead. *Nat. Rev. Drug Discov.* **19**, 93–111 (2020).
2. Gatta, G. et al. Rare cancers are not so rare: the rare cancer burden in Europe. *Eur. J. Cancer* **47**, 2493–2511 (2011).
3. Matsuda, T. et al. Rare cancers are not rare in Asia as well: the rare cancer burden in East Asia. *Cancer Epidemiol.* **67**, 101702 (2020).
4. National Cancer Institute. *NCI Dictionary of Cancer Terms* https://www.cancer.gov/publications/dictionaries/cancer-terms/def/rare-cancer (2022).
5. McCoach, C. E. & Doebele, R. C. The minority report: targeting the rare oncogenes in NSCLC. *Curr. Treat. Options Oncol.* **15**, 644–657 (2014).
6. Centers for Disease Control and Prevention. Annual trends in new cancers. https://gis.cdc.gov/Cancer/USCS/#/Trends/ (2022).
7. Guo, Y. et al. Recent progress in rare oncogenic drivers and targeted therapy for non-small cell lung cancer. *Onco Targets Ther.* **12**, 10343–10360 (2019).
8. Cancer Genome Atlas Research Network. Comprehensive molecular profiling of lung adenocarcinoma. *Nature* **511**, 543–550 (2014).
9. Siegel, R. L., Miller, K. D., Fuchs, H. E. & Jemal, A. Cancer statistics, 2022. *CA Cancer J. Clin.* **72**, 7–33 (2022).
10. Tong, J. H. et al. MET amplification and exon 14 splice site mutation define unique molecular subgroups of non-small cell lung carcinoma with poor prognosis. *Clin. Cancer Res.* **22**, 3048–3056 (2016).
11. Friedlaender, A. et al. EGFR and HER2 exon 20 insertions in solid tumours: from biology to treatment. *Nat. Rev. Clin. Oncol.* **19**, 51–69 (2022).
12. Hammerman, P. S. et al. Mutations in the DDR2 kinase gene identify a novel therapeutic target in squamous cell lung cancer. *Cancer Discov.* **1**, 78–89 (2011).
13. The AACR Project GENIE Consortium. AACR Project GENIE: powering precision medicine through an international consortium. *Cancer Discov.* **7**, 818–831 (2017).
14. Luo, J. et al. Overcoming KRAS-mutant lung cancer. *Am. Soc. Clin. Oncol. Educ. Book* **42**, 1–11 (2022).
15. Du, Z. et al. Structure–function analysis of oncogenic EGFR kinase domain duplication reveals insights into activation and a potential approach for therapeutic targeting. *Nat. Commun.* **12**, 1382 (2021).
16. Hunihan, L. et al. RASGRF1 fusions activate oncogenic RAS signaling and confer sensitivity to MEK inhibition. *Clin. Cancer Res.* **28**, 3091–3103 (2022).
17. Reddy, V. P. et al. *BRAF* fusions in clinically advanced non-small cell lung cancer: an emerging target for anti-*BRAF* therapies [abstract]. *J. Clin. Oncol.* **35** (Suppl. 15), 9072 (2017).
18. Nagasaka, M. & Ou, S. I. NRG1 and NRG2 fusion positive solid tumor malignancies: a paradigm of ligand-fusion oncogenesis. *Trends Cancer* **8**, 242–258 (2022).
19. Ou, S. I. et al. Identification of novel *CDH1-NRG2α* and *F11R-NRG2α* fusions in NSCLC plus additional novel *NRG2α* fusions in other solid tumors by whole transcriptome sequencing. *JTO Clin. Res. Rep.* **2**, 100132 (2021).
20. Costa, F. A. et al. Revealing the BRD4-NOTCH3 fusion: a novel hill in the cancer landscape. *Lung Cancer* **154**, 146–150 (2021).
21. Van, A. N. et al. Protein kinase C fusion proteins are paradoxically loss of function in cancer. *J. Biol. Chem.* **296**, 100445 (2021).
22. Liu, S. et al. The genomic characteristics of ALK fusion positive tumors in Chinese NSCLC patients. *Front. Oncol.* **10**, 726 (2020).
23. Schram, A. M., Chang, M. T., Jonsson, P. & Drilon, A. Fusions in solid tumours: diagnostic strategies, targeted therapy, and acquired resistance. *Nat. Rev. Clin. Oncol.* **14**, 735–748 (2017).
24. Hechtman, J. F. et al. Pan-Trk immunohistochemistry is an efficient and reliable screen for the detection of NTRK fusions. *Am. J. Surg. Pathol.* **41**, 1547–1551 (2017).
25. Wang, K. et al. FGFR1-ERK1/2-SOX2 axis promotes cell proliferation, epithelial-mesenchymal transition, and metastasis in FGFR1-amplified lung cancer. *Oncogene* **37**, 5340–5354 (2018).
26. Su, W. et al. ARAF protein kinase activates RAS by antagonizing its binding to RASGAP NF1. *Mol. Cell* **82**, 2443–2457.e7 (2022).
27. Kim, H. et al. Extrachromosomal DNA is associated with oncogene amplification and poor outcome across multiple cancers. *Nat. Genet.* **52**, 891–897 (2020).
28. Yu, H. A. et al. Analysis of tumor specimens at the time of acquired resistance to EGFR-TKI therapy in 155 patients with EGFR-mutant lung cancers. *Clin. Cancer Res.* **19**, 2240–2247 (2013).
29. Zhao, J. & Xia, Y. Targeting HER2 alterations in non-small-cell lung cancer: a comprehensive review. *JCO Precis. Oncol.* **4**, 411–425 (2020).
30. Zabransky, D. J. et al. HER2 missense mutations have distinct effects on oncogenic signaling and migration. *Proc. Natl Acad. Sci. USA* **112**, E6205–E6214 (2015).
31. Vyse, S. & Huang, P. H. Targeting EGFR exon 20 insertion mutations in non-small cell lung cancer. *Signal. Transduct. Target. Ther.* **4**, 5 (2019).
32. Neel, D. S. et al. Differential subcellular localization regulates oncogenic signaling by ROS1 kinase fusion proteins. *Cancer Res.* **79**, 546–556 (2019).
33. Guo, R. et al. MET-dependent solid tumours – molecular diagnosis and targeted therapy. *Nat. Rev. Clin. Oncol.* **17**, 569–587 (2020).
34. Zingg, D. et al. Truncated FGFR2 is a clinically actionable oncogene in multiple cancers. *Nature* **608**, 609–617 (2022).
35. Smith, H. W. et al. An ErbB2 splice variant lacking exon 16 drives lung carcinoma. *Proc. Natl Acad. Sci. USA* **117**, 20139–20148 (2020).
36. Haigis, K. M. KRAS alleles: the devil is in the detail. *Trends Cancer* **3**, 686–697 (2017).
37. Hayashi, T. et al. RASA1 and NF1 are preferentially co-mutated and define a distinct genetic subset of smoking-associated non-small cell lung carcinomas sensitive to MEK inhibition. *Clin. Cancer Res.* **24**, 1436–1447 (2018).
38. Castel, P. et al. RIT1 oncoproteins escape LZTR1-mediated proteolysis. *Science* **363**, 1226–1230 (2019).
39. Yao, Z. et al. Tumours with class 3 BRAF mutants are sensitive to the inhibition of activated RAS. *Nature* **548**, 234–238 (2017).
40. Noeparast, A. et al. CRAF mutations in lung cancer can be oncogenic and predict sensitivity to combined type II RAF and MEK inhibition. *Oncogene* **38**, 5933–5941 (2019).
41. Arcila, M. E. et al. MAP2K1 (MEK1) mutations define a distinct subset of lung adenocarcinoma associated with smoking. *Clin. Cancer Res.* **21**, 1935–1943 (2015).
42. French, C. NUT midline carcinoma. *Nat. Rev. Cancer* **14**, 149–150 (2014).
43. Wang, K. et al. PEST domain mutations in Notch receptors comprise an oncogenic driver segment in triple-negative breast cancer sensitive to a gamma-secretase inhibitor. *Clin. Cancer Res.* **21**, 1487–1496 (2015).
44. Jordan, E. J. et al. Prospective comprehensive molecular characterization of lung adenocarcinomas for efficient patient matching to approved and emerging therapies. *Cancer Discov.* **7**, 596–609 (2017).
45. Perez-Moreno, P., Brambilla, E., Thomas, R. & Soria, J. C. Squamous cell carcinoma of the lung: molecular subtypes and therapeutic opportunities. *Clin. Cancer Res.* **18**, 2443–2451 (2012).
46. Nishino, M. et al. Histologic and cytomorphologic features of ALK-rearranged lung adenocarcinomas. *Mod. Pathol.* **25**, 1462–1472 (2012).
47. Lee, S. E. et al. Comprehensive analysis of RET and ROS1 rearrangement in lung adenocarcinoma. *Mod. Pathol.* **28**, 468–479 (2015).
48. Nagasaka, M. & Ou, S. I. Neuregulin 1 fusion-positive NSCLC. *J. Thorac. Oncol.* **14**, 1354–1359 (2019).
49. Chang, J. C. et al. Comprehensive molecular and clinicopathologic analysis of 200 pulmonary invasive mucinous adenocarcinomas identifies distinct characteristics of molecular subtypes. *Clin. Cancer Res.* **27**, 4066–4076 (2021).
50. Shim, H. S. et al. Unique genetic and survival characteristics of invasive mucinous adenocarcinoma of the lung. *J. Thorac. Oncol.* **10**, 1156–1162 (2015).
51. Lee, J. K. et al. Characterization of non-small-cell lung cancers with MET exon 14 skipping alterations detected in tissue or liquid: clinicogenomics and real-world treatment patterns. *JCO Precis. Oncol.* https://doi.org/10.1200/PO.21.00122 (2021).
52. Socinski, M. A., Pennell, N. A. & Davies, K. D. MET exon 14 skipping mutations in non-small-cell lung cancer: an overview of biology, clinical outcomes, and testing considerations. *JCO Precis. Oncol.* https://doi.org/10.1200/PO.20.00516 (2021).
53. Al-Samkari, H. et al. Impact of ALK rearrangement on venous and arterial thrombotic risk in NSCLC. *J. Thorac. Oncol.* **15**, 1497–1506 (2020).
54. Ng, T. L. et al. ROS1 gene rearrangements are associated with an elevated risk of peridiagnosis thromboembolic events. *J. Thorac. Oncol.* **14**, 596–605 (2019).
55. Shaw, A. T. & Engelman, J. A. ALK in lung cancer: past, present, and future. *J. Clin. Oncol.* **31**, 1105–1111 (2013).
56. Tsuta, K. et al. RET-rearranged non-small-cell lung carcinoma: a clinicopathological and molecular analysis. *Br. J. Cancer* **110**, 1571–1578 (2014).
57. Parikh, D. A. et al. Characteristics of patients with ROS1+ cancers: results from the first patient-designed, global, pan-cancer ROS1 data repository. *JCO Oncol. Pract.* **16**, e183–e189 (2020).
58. Awad, M. M. et al. *MET* exon 14 mutations in non-small-cell lung cancer are associated with advanced age and stage-dependent *MET* genomic amplification and c-Met overexpression. *J. Clin. Oncol.* **34**, 721–730 (2016).
59. Riely, G. J. et al. Frequency and distinctive spectrum of KRAS mutations in never smokers with lung adenocarcinoma. *Clin. Cancer Res.* **14**, 5731–5734 (2008).
60. Camidge, D. R. et al. Anaplastic lymphoma kinase gene rearrangements in non-small cell lung cancer are associated with prolonged progression-free survival on pemetrexed. *J. Thorac. Oncol.* **6**, 774–780 (2011).
61. Drilon, A. et al. Clinical outcomes with pemetrexed-based systemic therapies in RET-rearranged lung cancers. *Ann. Oncol.* **27**, 1286–1291 (2016).
62. Chen, Y. F. et al. Efficacy of pemetrexed-based chemotherapy in patients with ROS1 fusion-positive lung adenocarcinoma compared with in patients harboring other driver mutations in East Asian populations. *J. Thorac. Oncol.* **11**, 1140–1152 (2016).
63. Marchetti, A. et al. Clinical features and outcome of patients with non-small-cell lung cancer harboring BRAF mutations. *J. Clin. Oncol.* **29**, 3574–3579 (2011).
64. Wang, Y. et al. Outcomes of pemetrexed-based chemotherapies in HER2-mutant lung cancers. *BMC Cancer* **18**, 326 (2018).

65. Naidoo, J. et al. Epidermal growth factor receptor exon 20 insertions in advanced lung adenocarcinomas: clinical outcomes and response to erlotinib. *Cancer* **121**, 3212–3220 (2015).
66. Rosen, E. Y. et al. TRK fusions are enriched in cancers with uncommon histologies and the absence of canonical driver mutations. *Clin. Cancer Res.* **26**, 1624–1632 (2020).
67. Drilon, A. et al. Clinicopathologic features and response to therapy of NRG1 fusion-driven lung cancers: the eNRGy1 global multicenter registry. *J. Clin. Oncol.* **39**, 2791–2802 (2021).
68. Mazieres, J. et al. Immune checkpoint inhibitors for patients with advanced lung cancer and oncogenic driver alterations: results from the IMMUNOTARGET registry. *Ann. Oncol.* **30**, 1321–1328 (2019).
69. Negrao, M. V. et al. Oncogene-specific differences in tumor mutational burden, PD-L1 expression, and outcomes from immunotherapy in non-small cell lung cancer. *J. Immunother. Cancer* https://doi.org/10.1136/jitc-2021-002891 (2021).
70. Guisier, F. et al. Brief report: First-line pembrolizumab in metastatic non-small cell lung cancer habouring MET exon 14 skipping mutation and PD-L1 ≥50% (GFPC 01-20 study). *Clin. Lung Cancer* **23**, e545–e549 (2022).
71. Schoenfeld, A. J. et al. Severe immune-related adverse events are common with sequential PD-(L)1 blockade and osimertinib. *Ann. Oncol.* **30**, 839–844 (2019).
72. Lin, J. J. et al. Increased hepatotoxicity associated with sequential immune checkpoint inhibitor and crizotinib therapy in patients with non-small cell lung cancer. *J. Thorac. Oncol.* **14**, 135–140 (2019).
73. McCoach, C. E. et al. Hypersensitivity reactions to selpercatinib treatment with or without prior immune checkpoint inhibitor therapy in patients with NSCLC in LIBRETTO-001. *J. Thorac. Oncol.* **17**, 768–778 (2022).
74. Yang, S. R. et al. Precision medicine in non-small cell lung cancer: current applications and future directions. *Semin. Cancer Biol.* **84**, 184–198 (2022).
75. Lindeman, N. I. et al. Molecular testing guideline for selection of lung cancer patients for EGFR and ALK tyrosine kinase inhibitors: guideline from the College of American Pathologists, International Association for the Study of Lung Cancer, and Association for Molecular Pathology. *J. Thorac. Oncol.* **8**, 823–859 (2013).
76. Lindeman, N. I. et al. Updated molecular testing guideline for the selection of lung cancer patients for treatment with targeted tyrosine kinase inhibitors: guideline from the College of American Pathologists, the International Association for the Study of Lung Cancer, and the Association for Molecular Pathology. *Arch. Pathol. Lab. Med.* **142**, 321–346 (2018).
77. Cheng, D. T. 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.* **17**, 251–264 (2015).
78. Passiglia, F., Malapelle, U., Normanno, N. & Pinto, C. Optimizing diagnosis and treatment of EGFR exon 20 insertions mutant NSCLC. *Cancer Treat. Rev.* **109**, 102438 (2022).
79. Drilon, A., Cappuzzo, F., Ou, S. I. & Camidge, D. R. Targeting MET in lung cancer: will expectations finally be MET? *J. Thorac. Oncol.* **12**, 15–26 (2017).
80. Davies, K. D. et al. Comparison of molecular testing modalities for detection of ROS1 rearrangements in a cohort of positive patient samples. *J. Thorac. Oncol.* **13**, 1474–1482 (2018).
81. Benayed, R. et al. High yield of RNA sequencing for targetable kinase fusions in lung adenocarcinomas with no mitogenic driver alteration detected by DNA sequencing and low tumor mutation burden. *Clin. Cancer Res.* **25**, 4712–4722 (2019).
82. Drilon, A. et al. Response to ERBB3-directed targeted therapy in NRG1-rearranged cancers. *Cancer Discov.* **8**, 686–695 (2018).
83. Yang, S. R. et al. A performance comparison of commonly used assays to detect RET fusions. *Clin. Cancer Res.* **27**, 1316–1328 (2021).
84. Poirot, B. et al. MET exon 14 alterations and new resistance mutations to tyrosine kinase inhibitors: risk of inadequate detection with current amplicon-based NGS panels. *J. Thorac. Oncol.* **12**, 1582–1587 (2017).
85. Davies, K. D. et al. DNA-based versus RNA-based detection of MET exon 14 skipping events in lung cancer. *J. Thorac. Oncol.* **14**, 737–741 (2019).
86. Guo, R. et al. MET exon 14-altered lung cancers and MET inhibitor resistance. *Clin. Cancer Res.* **27**, 799–806 (2021).
87. Desmeules, P. et al. Performance of an RNA-based next-generation sequencing assay for combined detection of clinically actionable fusions and hotspot mutations in NSCLC. *JTO Clin. Res. Rep.* **3**, 100276 (2022).
88. Cohen, D. et al. Optimizing mutation and fusion detection in NSCLC by sequential DNA and RNA sequencing. *J. Thorac. Oncol.* **15**, 1000–1014 (2020).
89. Hovelson, D. H. et al. Development and validation of a scalable next-generation sequencing system for assessing relevant somatic variants in solid tumors. *Neoplasia* **17**, 385–399 (2015).
90. Solomon, J. P. et al. Bioinformatically-expanded next-generation sequencing analysis optimizes identification of therapeutically relevant MET copy number alterations in >50,000 tumors. *Clin. Cancer Res.* https://doi.org/10.1158/1078-0432.CCR-22-1321 (2022).
91. Schrock, A. B. et al. Characterization of 298 patients with lung cancer harboring MET exon 14 skipping alterations. *J. Thorac. Oncol.* **11**, 1493–1502 (2016).
92. Wolf, J. et al. Capmatinib in MET exon 14-mutated or MET-amplified non-small-cell lung cancer. *N. Engl. J. Med.* **383**, 944–957 (2020).
93. Al-Kateb, H., Nguyen, T. T., Steger-May, K. & Pfeifer, J. D. Identification of major factors associated with failed clinical molecular oncology testing performed by next generation sequencing (NGS). *Mol. Oncol.* **9**, 1737–1743 (2015).
94. Leighl, N. B. et al. Clinical utility of comprehensive cell-free DNA analysis to identify genomic biomarkers in patients with newly diagnosed metastatic non-small cell lung cancer. *Clin. Cancer Res.* **25**, 4691–4700 (2019).
95. Brannon, A. R. et al. Enhanced specificity of clinical high-sensitivity tumor mutation profiling in cell-free DNA via paired normal sequencing using MSK-ACCESS. *Nat. Commun.* **12**, 3770 (2021).
96. Finkle, J. D. et al. Validation of a liquid biopsy assay with molecular and clinical profiling of circulating tumor DNA. *NPJ Precis. Oncol.* **5**, 63 (2021).
97. Woodhouse, R. et al. Clinical and analytical validation of FoundationOne Liquid CDx, a novel 324-gene cfDNA-based comprehensive genomic profiling assay for cancers of solid tumor origin. *PLoS ONE* **15**, e0237802 (2020).
98. Bale, T. A. et al. Clinical experience of cerebrospinal fluid-based liquid biopsy demonstrates superiority of cell-free DNA over cell pellet genomic DNA for molecular profiling. *J. Mol. Diagn.* **23**, 742–752 (2021).
99. Yang, S. R. et al. Targeted deep sequencing of cell-free DNA in serous body cavity fluids with malignant, suspicious, and benign cytology. *Cancer Cytopathol.* **128**, 43–56 (2020).
100. Negishi, R. et al. Transcriptomic profiling of single circulating tumor cells provides insight into human metastatic gastric cancer. *Commun. Biol.* **5**, 20 (2022).
101. Hasegawa, N. et al. Highly sensitive fusion detection using plasma cell-free RNA in non-small-cell lung cancers. *Cancer Sci.* **112**, 4393–4403 (2021).
102. Dunwell, T. L. et al. Adaptor template oligo-mediated sequencing (ATOM-Seq) is a new ultra-sensitive UMI-based NGS library preparation technology for use with cfDNA and cfRNA. *Sci. Rep.* **11**, 3138 (2021).
103. Wang, M. C. et al. Methods for collection of extracellular vesicles and their content RNA as liquid biopsy for lung cancer detection: application of differential centrifugation and annexin A5 coated beads. *Curr. Issues Mol. Biol.* **44**, 2374–2386 (2022).
104. Izumi, H. et al. The CLIP1-LTK fusion is an oncogenic driver in non-small-cell lung cancer. *Nature* **600**, 319–323 (2021).
105. Carrot-Zhang, J. et al. Whole-genome characterization of lung adenocarcinomas lacking the RTK/RAS/RAF pathway. *Cell Rep.* **34**, 108707 (2021).
106. Roskoski, R. Jr Properties of FDA-approved small molecule protein kinase inhibitors: a 2022 update. *Pharmacol. Res.* **175**, 106037 (2022).
107. Dar, A. C. & Shokat, K. M. The evolution of protein kinase inhibitors from antagonists to agonists of cellular signaling. *Annu. Rev. Biochem.* **80**, 769–795 (2011).
108. Yao, Z. et al. RAF inhibitor PLX8394 selectively disrupts BRAF dimers and RAS-independent BRAF-mutant-driven signaling. *Nat. Med.* **25**, 284–291 (2019).
109. Alabi, S. et al. Mutant-selective degradation by BRAF-targeting PROTACs. *Nat. Commun.* **12**, 920 (2021).
110. Li, J. W., Zheng, G., Kaye, F. J. & Wu, L. PROTAC therapy as a new targeted therapy for lung cancer. *Mol. Ther.* https://doi.org/10.1016/j.ymthe.2022.11.011 (2022).
111. Burslem, G. M. et al. The advantages of targeted protein degradation over inhibition: an RTK case study. *Cell Chem. Biol.* **25**, 67–77.e3 (2018).
112. Kinoshita, I. et al. A phase II study of trastuzumab monotherapy in pretreated patients with non-small cell lung cancers (NSCLCs) harboring HER2 alterations: HOT1303-B trial. *Ann. Oncol.* **29**, viii540 (2018).
113. Drago, J. Z., Modi, S. & Chandarlapaty, S. Unlocking the potential of antibody–drug conjugates for cancer therapy. *Nat. Rev. Clin. Oncol.* **18**, 327–344 (2021).
114. Ackerman, S. E. et al. Immune-stimulating antibody conjugates elicit robust myeloid activation and durable antitumor immunity. *Nat. Cancer* **2**, 18–33 (2021).
115. Li, B. T. et al. Trastuzumab deruxtecan in HER2-mutant non-small-cell lung cancer. *N. Engl. J. Med.* **386**, 241–251 (2021).
116. Camidge, D. R. et al. Telisotuzumab vedotin (Teliso-V) monotherapy in patients (pts) with previously treated c-Met–overexpressing (OE) advanced non-small cell lung cancer (NSCLC) [abstract]. *J. Clin. Oncol.* **40** (Suppl. 16), 9016 (2022).
117. Paik, P. K. et al. Tepotinib in non-small-cell lung cancer with MET exon 14 skipping mutations. *N. Engl. J. Med.* **383**, 931–943 (2020).
118. Planchard, D. et al. Dabrafenib plus trametinib in patients with previously untreated BRAFV600E-mutant metastatic non-small-cell lung cancer: an open-label, phase 2 trial. *Lancet Oncol.* **18**, 1307–1316 (2017).
119. Zhou, C. et al. Treatment outcomes and safety of mobocertinib in platinum-pretreated patients with EGFR exon 20 insertion-positive metastatic non-small cell lung cancer: a phase 1/2 open-label nonrandomized clinical trial. *JAMA Oncol.* **7**, e214761 (2021).
120. Park, K. et al. Amivantamab in EGFR exon 20 insertion-mutated non-small-cell lung cancer progressing on platinum chemotherapy: initial results from the CHRYSALIS phase I study. *J. Clin. Oncol.* **39**, 3391–3402 (2021).
121. Krebs, M. et al. Amivantamab in patients with NSCLC with MET exon 14 skipping mutation: updated results from the CHRYSALIS study [abstract]. *J. Clin. Oncol.* **40** (Suppl. 16), 9008 (2022).
122. Zhou, C. et al. Pyrotinib in HER2-mutant advanced lung adenocarcinoma after platinum-based chemotherapy: a multicenter, open-label, single-arm, phase II study. *J. Clin. Oncol.* **38**, 2753–2761 (2020).
123. Elamin, Y. Y. et al. Poziotinib for patients with HER2 exon 20 mutant non-small-cell lung cancer: results from a phase II trial. *J. Clin. Oncol.* **40**, 702–709 (2022).
124. Li, B. T. et al. Ado-trastuzumab emtansine for patients with HER2-mutant lung cancers: results from a phase II basket trial. *J. Clin. Oncol.* **36**, 2532–2537 (2018).
125. Mao, Z. et al. KRAS(G12D) can be targeted by potent inhibitors via formation of salt bridge. *Cell Discov.* **8**, 5 (2022).
126. Planchard, D. et al. Dabrafenib in patients with BRAF(V600E)-positive advanced non-small-cell lung cancer: a single-arm, multicentre, open-label, phase 2 trial. *Lancet Oncol.* **17**, 642–650 (2016).

127. Subbiah, V. et al. Efficacy of vemurafenib in patients with non-small-cell lung cancer with BRAF V600 mutation: an open-label, single-arm cohort of the histology-independent VE-BASKET study. *JCO Precis. Oncol.* **3**, PO.18.00266 (2019).
128. Berger, A. H. et al. Oncogenic RIT1 mutations in lung adenocarcinoma. *Oncogene* **33**, 4418–4423 (2014).
129. Moreno, V. et al. Extended follow-up of efficacy and safety of larotrectinib in patients with TRK fusion lung cancer [abstract EP08.02-148]. *J. Thorac. Oncol.* **17** (Suppl. 9), S473–S474 (2022).
130. Drilon, A. et al. ROS1-dependent cancers – biology, diagnostics and therapeutics. *Nat. Rev. Clin. Oncol.* **18**, 35–55 (2021).
131. Alexander, M. et al. A multicenter study of thromboembolic events among patients diagnosed with ROS1-rearranged non-small cell lung cancer. *Lung Cancer* **142**, 34–40 (2020).
132. Shaw, A. T. et al. Crizotinib in ROS1-rearranged advanced non-small-cell lung cancer (NSCLC): updated results, including overall survival, from PROFILE 1001. *Ann. Oncol.* **30**, 1121–1126 (2019).
133. Drilon, A. et al. Long-term efficacy and safety of entrectinib in ROS1 fusion-positive NSCLC. *JTO Clin. Res. Rep.* **3**, 100332 (2022).
134. Drilon, A. et al. Efficacy of selpercatinib in RET fusion-positive non-small-cell lung cancer. *N. Engl. J. Med.* **383**, 813–824 (2020).
135. Gainor, J. F. et al. Pralsetinib for RET fusion-positive non-small-cell lung cancer (ARROW): a multi-cohort, open-label, phase 1/2 study. *Lancet Oncol.* **22**, 959–969 (2021).
136. Solomon, B. J. et al. First-line crizotinib versus chemotherapy in ALK-positive lung cancer. *N. Engl. J. Med.* **371**, 2167–2177 (2014).
137. Soria, J.-C. et al. First-line ceritinib versus platinum-based chemotherapy in advanced *ALK*-rearranged non-small-cell lung cancer (ASCEND-4): a randomised, open-label, phase 3 study. *Lancet* **389**, 917–929 (2017).
138. Peters, S. et al. Alectinib versus crizotinib in untreated *ALK*-positive non-small-cell lung cancer. *N. Engl. J. Med.* **377**, 829–838 (2017).
139. Mok, T. et al. Updated overall survival and final progression-free survival data for patients with treatment-naive advanced *ALK*-positive non-small-cell lung cancer in the ALEX study. *Ann. Oncol.* **31**, 1056–1064 (2020).
140. Camidge, D. R. et al. Brigatinib versus crizotinib in ALK-positive non-small-cell lung cancer. *N. Engl. J. Med.* **379**, 2027–2039 (2018).
141. Shaw, A. T. et al. First-line lorlatinib or crizotinib in advanced ALK-positive lung cancer. *N. Engl. J. Med.* **383**, 2018–2029 (2020).
142. Shaw, A. T. et al. Lorlatinib in non-small-cell lung cancer with ALK or ROS1 rearrangement: an international, multicentre, open-label, single-arm first-in-man phase 1 trial. *Lancet Oncol.* **18**, 1590–1599 (2017).
143. Schram, A. M. et al. Efficacy and safety of zenocutuzumab, a HER2 x HER3 bispecific antibody, across advanced NRG1 fusion (NRG1+) cancers [abstract]. *J. Clin. Oncol.* **40** (Suppl. 16), 105 (2022).
144. Carrizosa, D. R. et al. CRESTONE: initial efficacy and safety of seribantumab in solid tumors harboring *NRG1* fusions [abstract]. *J. Clin. Oncol.* **40** (Suppl. 16), 3006 (2022).
145. Janne, P. A. et al. Efficacy and safety of patritumab deruxtecan (HER3-DXd) in EGFR inhibitor-resistant, EGFR-mutated non-small cell lung cancer. *Cancer Discov.* **12**, 74–89 (2022).
146. Davies, K. D. et al. Dramatic response to crizotinib in a patient with lung cancer positive for an HLA-DRB1-MET gene fusion. *JCO Precis. Oncol.* https://doi.org/10.1200/PO.17.00117 (2017).
147. Qin, A. et al. Detection of known and novel FGFR fusions in non-small cell lung cancer by comprehensive genomic profiling. *J. Thorac. Oncol.* **14**, 54–62 (2019).
148. Cooper, A. J. et al. Identification of a RAS-activating TMEM87A-RASGRF1 fusion in an exceptional responder to sunitinib with non-small cell lung cancer. *Clin. Cancer Res.* **26**, 4072–4079 (2020).
149. Camidge, D. R. et al. Crizotinib in patients with MET-amplified NSCLC. *J. Thorac. Oncol.* **16**, 1017–1029 (2021).
150. Le, X. et al. Tepotinib in patients (pts) with advanced non-small cell lung cancer (NSCLC) with *MET* amplification (*MET*amp) [abstract]. *J. Clin. Oncol.* **39** (Suppl. 15), 9021 (2021).
151. Hainsworth, J. D. et al. Targeted therapy for advanced solid tumors on the basis of molecular profiles: results from MyPathway, an open-label, phase IIa multiple basket study. *J. Clin. Oncol.* **36**, 536–542 (2018).
152. Song, Z. et al. Pyrotinib in patients with HER2-amplified advanced non-small cell lung cancer: a prospective, multicenter, single-arm trial. *Clin. Cancer Res.* **28**, 461–467 (2022).
153. Drilon, A. et al. Cabozantinib in patients with advanced RET-rearranged non-small-cell lung cancer: an open-label, single-centre, phase 2, single-arm trial. *Lancet Oncol.* **17**, 1653–1660 (2016).
154. Lee, S. H. et al. Vandetanib in pretreated patients with advanced non-small cell lung cancer-harboring RET rearrangement: a phase II clinical trial. *Ann. Oncol.* **28**, 292–297 (2017).
155. Yakes, F. M. et al. Cabozantinib (XL184), a novel MET and VEGFR2 inhibitor, simultaneously suppresses metastasis, angiogenesis, and tumor growth. *Mol. Cancer Ther.* **10**, 2298–2308 (2011).
156. Demetri, G. D. et al. Updated integrated analysis of the efficacy and safety of entrectinib in patients with NTRK fusion-positive solid tumors. *Clin. Cancer Res.* **28**, 1302–1312 (2022).
157. Yu, H. A. et al. Phase (Ph) 1/2a study of CLN-081 in patients (pts) with NSCLC with EGFR exon 20 insertion mutations (Ins20) [abstract]. *J. Clin. Oncol.* **40** (Suppl. 16), 9007 (2022).
158. Drilon, A. et al. Antitumor activity of crizotinib in lung cancers harboring a MET exon 14 alteration. *Nat. Med.* **26**, 47–51 (2020).
159. Patil, T. et al. The incidence of brain metastases in stage IV ROS1-rearranged non-small cell lung cancer and rate of central nervous system progression on crizotinib. *J. Thorac. Oncol.* **13**, 1717–1726 (2018).
160. Shaw, A. T. et al. Lorlatinib in advanced ROS1-positive non-small-cell lung cancer: a multicentre, open-label, single-arm, phase 1-2 trial. *Lancet Oncol.* **20**, 1691–1701 (2019).
161. Bartsch, R. et al. Trastuzumab deruxtecan in HER2-positive breast cancer with brain metastases: a single-arm, phase 2 trial. *Nat. Med.* **28**, 1840–1847 (2022).
162. Shu, C. A. et al. Amivantamab and lazertinib in patients with EGFR-mutant non-small cell lung (NSCLC) after progression on osimertinib and platinum-based chemotherapy: updated results from CHRYSALIS-2 [abstract]. *J. Clin. Oncol.* **40** (Suppl. 16), 9006 (2022).
163. Aragon-Sanabria, V. et al. Ultrasmall nanoparticle delivery of doxorubicin improves therapeutic index for high-grade glioma. *Clin. Cancer Res.* **28**, 2938–2952 (2022).
164. Cocco, E. et al. TRK xDFG mutations trigger a sensitivity switch from type I to II kinase inhibitors. *Cancer Discov.* **11**, 126–141 (2021).
165. Murray, B. W. et al. TPX-0131, a potent CNS-penetrant, next-generation inhibitor of wild-type ALK and ALK-resistant mutations. *Mol. Cancer Ther.* **20**, 1499–1507 (2021).
166. Pelish, H. E. et al. NUV-655 (NVL-655) is a selective, brain-penetrant ALK inhibitor with antitumor activity against the lorlatinib-resistant G1202R/L1196M compound mutation [abstract]. *Cancer Res.* **81** (Suppl. 13), 1468 (2021).
167. Yun, M. R. et al. Repotrectinib exhibits potent antitumor activity in treatment-naïve and solvent-front-mutant ROS1-rearranged non-small cell lung cancer. *Clin. Cancer Res.* **26**, 3287–3295 (2020).
168. Rotow, J. et al. Combination osimertinib plus selpercatinib for EGFR-mutant non-small cell lung cancer (NSCLC) with acquired RET fusions [abstract FP14.07]. *J. Thorac. Oncol.* **16** (Suppl. 3), S230 (2021).
169. Dolgin, E. Oncogene-specific advocacy groups bring a patient-centric perspective to studies of lung cancer. *Nature* **587**, S16–S17 (2020).
170. Global ROS1 Initiative. *The ROS1ders* https://www.theros1ders.org/global-ros1-initiative (accessed 10 February 2023).
171. Maemondo, M. et al. Gefitinib or chemotherapy for non-small-cell lung cancer with mutated EGFR. *N. Engl. J. Med.* **362**, 2380–2388 (2010).
172. Kim, E. S. et al. The BATTLE trial: personalizing therapy for lung cancer. *Cancer Discov.* **1**, 44–53 (2011).
173. Herbst, R. S. et al. Lung master protocol (Lung-MAP) – a biomarker-driven protocol for accelerating development of therapies for squamous cell lung cancer: SWOG S1400. *Clin. Cancer Res.* **21**, 1514–1524 (2015).
174. Middleton, G. et al. The National Lung Matrix trial of personalized therapy in lung cancer. *Nature* **583**, 807–812 (2020).
175. Hyman, D. M., Taylor, B. S. & Baselga, J. Implementing genome-driven oncology. *Cell* **168**, 584–599 (2017).
176. Farago, A. F. et al. Clinicopathologic features of non-small-cell lung cancer harboring an NTRK gene fusion. *JCO Precis. Oncol.* https://doi.org/10.1200/po.18.00037 (2018).
177. Yang, J. C. H. et al. Rationale and design of ON-TRK: a novel prospective non-interventional study in patients with TRK fusion cancer treated with larotrectinib. *BMC Cancer* **22**, 625 (2022).
178. Hobbs, B. P. et al. Seamless designs: current practice and considerations for early-phase drug development in oncology. *J. Natl Cancer Inst.* **111**, 118–128 (2019).
179. Bhatt, D. L. & Mehta, C. Adaptive designs for clinical trials. *N. Engl. J. Med.* **375**, 65–74 (2016).
180. Vokinger, K. N. & Kesselheim, A. S. Application of orphan drug designation to cancer treatments (2008–2017): a comprehensive and comparative analysis of the USA and EU. *BMJ Open.* **9**, e028634 (2019).
181. US Food & Drug Administration. Fast track, breakthrough therapy, accelerated approval, priority review. *FDA* https://www.fda.gov/patients/learn-about-drug-and-device-approvals/fast-track-breakthrough-therapy-accelerated-approval-priority-review (2018).
182. Kloda, J. H. & Somerville, S. FDA's expedited review process: the need for speed. *appliedclinicaltrialsonline.com*, https://www.appliedclinicaltrialsonline.com/view/fda-s-expedited-review-process-need-speed (11 March 2015).
183. Chodankar, D. Introduction to real-world evidence studies. *Perspect. Clin. Res.* **12**, 171–174 (2021).
184. Gautschi, O. et al. Targeting RET in patients with RET-rearranged lung cancers: results from the global, multicenter RET registry. *J. Clin. Oncol.* **35**, 1403–1410 (2017).
185. US Food & Drug Administration. Framework for FDA's real-world evidence program. *FDA* https://www.fda.gov/media/120060/download (2018).
186. Elamin, Y. Y. et al. Poziotinib for *EGFR* exon 20-mutant NSCLC: clinical efficacy resistance mechanisms and impact of insertion location on drug sensitivity. *Cancer Cell* **40**, 754–767.e6 (2022).
187. Mazieres, J. et al. Combination of trastuzumab, pertuzumab, and docetaxel in patients with advanced non-small-cell lung cancer harboring HER2 mutations: results from the IFCT-1703 R2D2 trial. *J. Clin. Oncol.* **40**, 719–728 (2022).
188. Huang, X. et al. The efficacy of ado-trastuzumab emtansine in patients with ERBB2-aberrant non-small cell lung cancer: a systematic review. *Transl. Cancer Res.* **9**, 4507–4516 (2020).
189. Lu, S. et al. Phase II study of savolitinib in patients (pts) with pulmonary sarcomatoid carcinoma (PSC) and other types of non-small cell lung cancer (NSCLC) harboring MET exon 14 skipping mutations (METex14+) [abstract]. *J. Clin. Oncol.* **38** (Suppl. 15), 9519 (2020).
190. Horn, L. et al. Ensartinib vs crizotinib for patients with anaplastic lymphoma kinase-positive non-small cell lung cancer: a randomized clinical trial. *JAMA Oncol.* **7**, 1617–1625 (2021).

191. Lim, S. M. et al. Open-label, multicenter, phase II study of ceritinib in patients with non-small-cell lung cancer harboring ROS1 rearrangement. *J. Clin. Oncol.* **35**, 2613–2618 (2017).
192. Turning Point Therapeutics. Turning Point Therapeutics announces positive topline data by blinded independent central review for repotrectinib across all ROS1-positive NSCLC cohorts of phase 1/2 TRIDENT-1 study. *Turning Point Therapeutics* https://www.tptherapeutics.com/news-releases/news-release-details/turning-point-therapeutics-announces-positive-topline-data/ (2022).
193. Sanchez-Vega, F. et al. Oncogenic signaling pathways in the cancer genome atlas. *Cell* **173**, 321–337.e10 (2018).
194. Chen, J. et al. Genomic landscape of lung adenocarcinoma in East Asians. *Nat. Genet.* **52**, 177–186 (2020).
195. Cerami, E. et al. The cBio cancer genomics portal: an open platform for exploring multidimensional cancer genomics data. *Cancer Discov.* **2**, 401–404 (2012).
196. Gao, J. et al. Integrative analysis of complex cancer genomics and clinical profiles using the cBioPortal. *Sci. Signal.* **6**, pl1 (2013).
197. ASCO.org. Lung cancer - non-small cell: statistics. *Cancer.Net* https://www.cancer.net/cancer-types/lung-cancer-non-small-cell/statistics (2022).
198. Chang, M. T. et al. Identifying recurrent mutations in cancer reveals widespread lineage diversity and mutational specificity. *Nat. Biotechnol.* **34**, 155–163 (2016).
199. Hida, T. et al. A phase 2 study of lenvatinib in patients with RET fusion-positive lung adenocarcinoma. *Lung Cancer* **138**, 124–130 (2019).

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Competing interests
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Additional information
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