Review

Patient Selection Approaches in FGFR Inhibitor Trials—Many Paths to the Same End?

Peter Ellinghaus 1, Daniel Neureiter 2,3, Hendrik Nogai 4, Sebastian Stintzing 5 and Matthias Ocker 5,6,*

1 Global Clinical Development Oncology, Merck Healthcare KGaA, 64293 Darmstadt, Germany
2 Institute of Pathology, University Clinics Salzburg, Paracelsus Medical University, 5020 Salzburg, Austria
3 Cancer Cluster Salzburg, 5020 Salzburg, Austria
4 Ryvu Therapeutics, 30-394 Krakow, Poland
5 Division of Hematology, Oncology, and Tumor Immunology (Campus Charité Mitte), Medical Department, Charité University Medicine Berlin, 10117 Berlin, Germany
6 Anji Pharmaceuticals, Cambridge, MA 02142, USA
* Correspondence: matthias.ocker@charite.de

Abstract: Inhibitors of fibroblast growth factor receptor (FGFR) signaling have been investigated in various human cancer diseases. Recently, the first compounds received FDA approval in biomarker-selected patient populations. Different approaches and technologies have been applied in clinical trials, ranging from protein (immunohistochemistry) to mRNA expression (e.g., RNA in situ hybridization) and to detection of various DNA alterations (e.g., copy number variations, mutations, gene fusions). We review, here, the advantages and limitations of the different technologies and discuss the importance of tissue and disease context in identifying the best predictive biomarker for FGFR targeting therapies.

Keywords: fibroblast growth factor receptor; amplification; mutation; fusion; FGFR inhibitor; predictive biomarker; clinical trials

1. Introduction

Growth factor signaling has been identified as a major hallmark of cancer, leading to dysregulation of survival, growth and metabolic pathways [1]. Aberrant growth factor signaling via, e.g., mutations in tyrosine kinase domains, or overexpression or amplification of cognate receptors has recently also been linked to modulation of tumor microenvironment and, thus, plays a major role in controlling immune response against cancers [2,3].

Consequently, these pathways were identified early on as potential targets for cancer therapy by either inhibiting downstream signaling (e.g., alpelisib or copanlisib targeting phosphoinositide-3-kinase (PI3K), temsirolimus and other rapalogs targeting mammalian/mechanistic target of rapamycin (mTOR) or trametinib targeting mitogen-activated protein kinase kinase (MAPK/MEK)) or by directly interfering with upstream receptors. Seminal insights into growth factor receptor blockade were obtained from cetuximab and panitumumab targeting epithelial growth factor receptor (EGFR), trastuzumab targeting human epidermal growth factor receptor 2 (Her2/neu, erb-B2) and bevacizumab targeting vascular endothelial growth factor (VEGF). For the first two targets, a clear predictive biomarker selection technique was developed, ranging from simple target expression to downstream mutational profiles [4,5]. However, no such strategy is currently available for anti-angiogenic agents [6].

Recently, also, the fibroblast growth factor receptor (FGFR) family has entered the focus of drug development and the first compounds, erdafitinib (JNJ-42756493, Balversa™), pemigatinib (INCB054828, Pemazyre™), infgratinib (BGJ398, Truseltiq™) and, most recently, futibatinib (TAS-120, Lytgoib™), have received United States Food and Drug Administration (US FDA) approval for treatment of urothelial (bladder) cancer and intrahepatic...
biliary tract cancer, respectively [7,8]. While these US FDA-approved compounds use FGFR gene fusions and mutations as a companion diagnostic for patient selection, other agents use FGFR gene amplifications or FGFR overexpression to select patients in their respective clinical trials [9–11]. In this article, we will summarize and review the different approaches used for identification of patients for small-molecule FGFR inhibitor clinical trials in various cancer indications.

2. Brief Introduction to FGFR Biology and Signaling

Five fibroblast growth factor receptors (FGFRs) have been identified in humans, with four of them belonging to the family of transmembrane receptor tyrosine kinases and contain three immunoglobulin-like extracellular domains, mediating ligand specificity, a transmembrane spanning domain and an intracellular tyrosine kinase domain [12–15]. Twenty-two ligands for FGFRs have so far been identified in humans. They are usually subclustered into intracrine, endocrine and paracrine members. Except for intracrine fibroblast growth factor ligands (FGFs; FGF11–FGF14) that signal via the canonical receptor pathways, each FGF can bind to multiple FGFRs, leading to a complex interaction map of ligands and receptors (for more detail, see our previous review [16]). Ligand binding leads to receptor dimerization and subsequent phosphorylation of downstream signaling molecules (mostly fibroblast growth factor receptors substrate 2α, FRS2α, and growth factor receptor-bound protein 2, GRB2) into the PI3K-AKT or the RAS-RAF-MAPK pathway, which regulate cellular survival and growth mechanisms [17–19]. Other downstream targets include signal transducer and activator of transcription (STAT) molecules [20], adhesion molecules, such as N-cadherin [21–23] or the WNT/β-catenin pathway, thus, linking FGFR to invasion and metastasis formation [24–26] as well as chemoresistance via epithelial-mesenchymal transition [27,28] (Figure 1). Unlike FGFR1-3, FGFR4 does not have splice variants in IgIII, which generates the IIIb and IIIc transcript variants encoding different receptor isoforms. The lack of alternative splicing of IgIII reduces the ability to switch ligand binding specificity [29,30]. FGFR4 contains a unique amino acid (Cystein at position 522, Cys522) in the kinase domain, which is not present in FGFR1–3. Cys552 is conserved in just five other human protein kinases, including MK2, MK3, S6K2, STK40 and TTK. Thus, covalently targeting the Cys552 in FGFR4 is an appealing strategy for achieving selective inhibition of FGFR4, both with respect to isoform and kinome selectivity. This structural difference enables the design of FGFR4-specific inhibitors [31]. Selective inhibitors of FGFR4 have demonstrated clinical benefit in HCC patients with high FGF19 expression [32].

Overall, genomic alterations in FGFRs have been identified in approximately 6–7% of all human cancers [33–35]. The oncogenic potential of FGFRs has been linked to increased expression due to gene amplification (up to 66%), gene fusion/translocation (up to 8%) or gene mutations (up to 26%) in the signaling domains or abnormality of FGFR ligands [36–38]. It is interesting that these alterations show a distinct prevalence pattern in various human tumor types. While the frequency of FGFR2 mutations is highest in endometrial cancer (10–12%), it is below 5% in, e.g., non-small-cell lung cancer (NSCLC), gastric and urothelial cancer. Yet, FGFR3 mutations reach a prevalence of 75% in non-muscle-invasive bladder cancer, while it drops to 15% in the more aggressive muscle-invasive subtype [34,39–41]. Similar findings were observed for amplifications, which are rare for FGFR3 and FGFR4, while FGFR1 amplification was found in up to 19% of NSCLC and hormone-receptor-positive breast cancer (dropping to 4% in triple-negative breast cancer) [42–45]. Amplification of FGFR2 was found in up to 10% of gastric cancers [46,47] and FGFR2 gene fusions in up to 20% of intrahepatic cholangiocarcinomas and up to 6% of urothelial carcinomas [35]. Most prominent FGFR gene fusions represent FGFR3-TACC3 (transforming acidic colle-coil containing protein 3) in urothelial cancer and FGFR2-BICC1 (Bicc family binding protein 1) or FGFR2-AHCYL1 (adenosylhomocysteinase like 1) in intrahepatic cholangiocarcinomas [48–51]. These results indicate that expression of and genetic alterations in the various FGFR isoforms are highly context dependent and that a
A thorough understanding of the (dys-)regulated FGFR pathway is important to understand the optimal therapeutic setting for FGFR inhibitors. Recent data also suggest epigenetic mechanisms, such as methylation or miRNA expression, to regulate FGFR expression per se or in response to treatment, thus, representing a resistance mechanism [52–54].

Figure 1. Schematic representation of FGFR signaling and impact of various alterations. (A) Physiologic signaling upon ligand binding leads to various downstream signaling cascades affecting cellular survival, growth, migration, metabolism and interaction with cellular microenvironment. (B) Point mutations (marked in red) lead to constitutive activation by either affecting the extracellular ligand-binding domain or the intracellular tyrosine kinase domains. Signaling becomes independent of FGF ligand binding. (C) Gene fusions, rearrangements or translocations on DNA level (marked in dark green) lead to ligand-independent constitutive activation of the kinase domains by adding alternative kinase elements. (D) Gene amplification by DNA copy number alterations leads to higher expression of the receptor, providing more opportunities for ligands to bind and to activate the signaling cascade. It is noteworthy that all shown alterations also lead to increased mRNA expression levels but not all alterations lead to receptor overexpression. AKT: synonymous Protein Kinase B; β-cat: β-catenin; FGFR: fibroblast growth factor receptor; JAK: janus kinase; MAPK: mitogen-activated protein kinase; mTOR: mammalian/mechanistic target of rapamycin; PI3K: phosphoinositide-3-kiase; RAF: rapidly accelerated fibrosarcoma; RAS: rat sarcoma; STAT: signal transducer and activator of transcription; WNT: wingless and Int-1.

It was demonstrated that FGFR2 is able to induce expression of programmed cell death 1 ligand 1 (PD-L1) via the janus kinase (JAK)/STAT pathway in colorectal cancer [55] and a non-T-cell-inflamed phenotype was observed in FGFR3-driven urothelial cancer [56,57], although a recent study from Denmark could not confirm this finding [58]. Yet, preclinical data clearly demonstrate that inhibition of FGFR enhances the infiltration of CD8+ T
cells and inhibits tumor growth via modulation of the tumor microenvironment [59–61]. Wu et al. observed that the FGFR inhibitor-mediated blockade of the mitogen-activated protein kinase (MAPK)/extracellular-signal-regulated kinase (ERK) pathway in cancer-associated fibroblasts leads to diminished proliferation, migration and secretion of the vascular adhesion molecule 1 (VCAM-1) in these cells, which promotes T cell infiltration by breaking down the tumor/stroma barrier [62]. Furthermore, FGFR tyrosine kinase inhibitors were able to upregulate major histocompatibility complex (MHC) class I and class II expression via induction of the MHC Class II gene master regulator Class II transactivator (CIITA) and subsequent inhibition of MAPK and to augment the antitumor effects of FGFR1-reactive T cells [63]. Overall, the non-canonical effects of FGFR inhibitors in regulating the immune phenotype of tumors is still poorly understood and needs more experimental and clinical studies.

For more details on the metabolic pathways affected by FGFR signaling in the liver, we refer to other recently published reviews [64,65].

3. FGFR Inhibitors in Clinical Trials

This article focuses on small-molecule inhibitors of FGFR signaling, although other modalities, such as FGF ligand traps (e.g., GSK3052230), FGFR2-targeting antibody drug conjugates (e.g., aprutumab ixadotin/BAY 1187982) or receptor-blocking antibodies (e.g., bemarituzumab), have also been explored in early clinical trials [41].

Most of the small-molecule FGFR inhibitors (Table 1) are ATP-competitive inhibitors of several FGFR isoforms. Commonly, FGFR1–3 are inhibited at low nanomolar concentrations in a biochemical assay, while inhibition of FGFR4 is often less potent. Selective inhibitors of FGFR4, such as fisogatinib, roblitinib or H3B-6527, are irreversible covalent inhibitors, as is futibatinib that represents the only irreversible pan-FGFR inhibitor. Allosteric inhibitors, such as Alofanib (RPT835) or SSR128129E, have not yet reported human clinical data [66,67]. For many of the compounds listed in Table 1, clinical trials are still ongoing (please see [41], also for details on response rates and www.clinicaltrials.gov for more information, last accessed on 3 October 2022). So far, only erdafitinib, infigratinib, pemigatinib and futibatinib have received FDA approval for treatment of bladder cancer or intrahepatic cholangiocarcinoma.

The majority of the FGFR inhibitors that were used in clinical trials represent pan-FGFR inhibitors that usually target FGFR1, FGFR2 and FGFR3 at low-nanomolar IC50 values and FGFR4 at slightly higher values. Most of the compounds used a non-biomarker-selected all-comer population for dose escalation and switched to a distinct patient population for dose expansion and later phases of development. The tumor-agnostic potential of FGFR inhibitors, regardless of the underlying FGFR subtype altered, was recently demonstrated in the RAGNAR clinical trial with erdafitinib [68], in NCI-MATCH trial EAY131 with AZD4547 and in a large phase 1 study with rogaratinib [9]. Taken together, responses were observed in over 25 different malignancies. Interestingly, only the FGFR4-selective inhibitors explore the ligand FGF19 as a potential patient selection biomarker, while all other compounds focus on FGFR alterations, such as gene fusions, amplifications or mutations. As outlined above, these alterations show differential patterns in various human tumor types. Thus, assays for prospective patient selection either need a broad specificity or multiple assays need to be applied.

In the following section, different assays used in clinical development or as a full companion diagnostics tools for FGFR inhibitors will be discussed.
Table 1. Clinically tested FGFR inhibitors.

| Name                          | Selectivity | Indications                  | Phase | Biomarker                                                                 | Reference |
|-------------------------------|-------------|------------------------------|-------|----------------------------------------------------------------------------|-----------|
| ASP5878                       | FGFR1-4     | UC, HCC, sqLC                | 1     | FGFR3 fusion or mutation by FISH or PCR (UC), FGF19 overexp (HCC) or FGFR1 overexp (sqLC) by IHC | [69,70]  |
| AZD4547                       | FGFR1-4     | BC, GC, sqLC, agnostic       | 3     | FGFR copy number in ctDNA (BC), FISH (GC, sqLC), any FGFR alteration by NGS in the indication agnostic setting | [11,71–74] |
| Debio 1347                    | FGFR1-3     | Advanced solid tumors        | 1/2   | FISH, NGS                                                                  | [75,76]  |
| Derazantinib (ARQ087)         | FGFR1-3     | ihCC                         | 1/2   | FGFR2 fusion by FISH or NGS                                               | [77,78]  |
| Dovitinib (TKI-258)           | FGFR1 & 3   | RCC and other solid tumors   | 3     | No specific biomarker used                                                 | [79,80]  |
| E7090                         | FGFR1-3     | GC, ihCC, advanced solid tumors | 1/2 | FGFR2 amp (GC), FGFR2 fusion (ihCC), NGS                                  | [81–83]  |
| Erdafitinib (JNJ-42756493)    | FGFR1-4     | UC                           | approved | FGFR2/3 alterations by qRT-PCR                                           | [84,85]  |
| Fisogatinib (BLU-554)         | FGFR4       | HCC                          | 1/2   | FGF19 by IHC                                                               | [32]     |
| Futibatinib (TAS-120)         | FGFR1-4     | ihCC, GC, advanced solid tumors | approved | FGFR2 amp (GC), various FGFR aberrations                                 | [86–89]  |
| Infirgratinib (BGFR3098)      | FGFR1-3     | ihCC, gliomas                | approved | Any alteration of FGFR1 or FGFR3 (gliomas) or FGFR2 (ihCC)              | [90–92]  |
| LY2874455                     | FGFR1-4     | GC, NSCLC                    | 1     | FGFR1 amp (NSCLC), FGFR2 amp (GC)                                        | [93,94]  |
| ODM-203                       | FGFR1-4     | Advanced solid tumors        | 1     | Any genetic FGFR aberration                                               | [95,96]  |
| Pemigatinib (INCB054828)      | FGFR1-3     | ihCC                         | approved | NGS                                                                       | [97,98]  |
| Ponatinib                     | FGFR1-4     | ihCC                         | 3     | FGFR2 fusion/rearrangement by FISH or NGS                                | [99,100] |
| Roblitinib (FGFR401)          | FGFR4       | HCC                          | 1/2   | FGFR4 expression by PCR                                                   | [101]    |
| Rogaratinib (BAY 1163877)     | FGFR1-4     | Advanced solid tumors        | 1/2   | mRNA expression (RNA-ISH, Nanostring)                                    | [9,102,103] |

Amp: amplification; BC: breast cancer; FISH: fluorescence in situ hybridization; GC: gastric cancer; HCC: hepatocellular carcinoma; ihCC: intrahepatic cholangiocarcinoma; IHC: immunohistochemistry; NGS: next generation sequencing; NSCLC: non-small cell lung cancer; PCR: polymerase chain reaction; RCC: renal cell cancer; sqLC: squamous lung cancer; UC: urothelial carcinoma.
4. Predictive Biomarkers for FGFR Inhibitors

Early clinical trials with small-molecule FGFR inhibitors focused solely on histological tumor entities with a reasonable frequency of FGFR DNA alterations. FGFR1 amplification is the most frequently observed DNA abnormality in the squamous subtype of NSCLC (up to 20%). Meanwhile, it became obvious that not all NSCLC patients with an FGFR copy number gain had higher FGFR expression levels [104]. In gastric cancer, it could be shown that only homogenous FGFR2 gene amplification led to FGFR2 overexpression and, thus, to a treatment benefit [105]. A weak overlap between FGFR1 amplification and FGFR1 overexpression was also described in a large cohort of head and neck squamous cell carcinoma (HNSCC) patients [106].

Given the mode of action of small-molecule FGFR kinase inhibitors, the enzymatic activity can only be inhibited if a) there is a higher FGFR expression level within the tumor or b) if the enzymatic activity is increased by an activating single-nucleotide variant within the kinase domain. Very interestingly, in urothelial carcinoma, where FGFR3-activating mutations are most frequent (see before), it could be shown that the presence of a point mutation leads to strong overexpression of the mutant protein. Similar findings were observed for FGFR3 translocations in urothelial carcinoma and for FGFR2 fusions in cholangiocarcinoma, where the lack of the 3' end of the FGFR transcript being fused to another partner gene delays the microRNA-mediated degradation of the fusion transcript and, thus, increases FGFR fusion gene tumor expression levels accordingly. Thus, the selection of patients eligible for FGFR inhibitor therapy can be referred back to the degree of the FGFR overexpression within the tumor, surprisingly, even including overexpression of FGFR due to activating point mutations. For urothelial cancer, it has been known for decades that some tumors, especially in earlier stages of the disease, reveal FGFR overexpression without an underlying FGFR DNA alteration [107–109], e.g., an activating FGFR3 mutation or an FGFR3 gene fusion. It remained a matter of speculation if patients without such a DNA alteration may also benefit from FGFR inhibitor therapy. Schuler et al. demonstrated FGFR inhibitor sensitivity in FGFR-overexpressing urothelial carcinoma patients in the absence of a detectable FGFR DNA alteration [9]. A recent Phase 2 study in gastric cancer with the FGFR2-targeting monoclonal antibody bemarituzumab demonstrated FGFR2 overexpression via immunohistochemistry in 30% of screened patients, while other datasets suggest that the amplification of FGFR2 in tissue reaches 2.2–4% and 7.7% via ctDNA analysis [110,111]. Patients with overexpression of FGFR2b, even without ctDNA amplification, demonstrated a benefit from the addition of bemarituzumab to mFOLFOX6, supporting further evaluation of bemarituzumab in tumors with FGFR2b overexpression, regardless of an underlying FGFR2 gene amplification.

Recent data from the indication-agnostic NCI-MATCH Trial EAY131 (subprotocol W) also confirmed that alterations in FGFR, as detected by next-generation sequencing (NGS), in that tumor tissue is more common across a broad range of tumor diseases than previously expected. Here, various alterations were found also in, e.g., salivary gland tumors, rectal cancer, pancreatic cancer, prostate cancer and other tumors that were considered not to harbor FGFR alterations [74]. These findings corroborate the results from Schuler et al., who could demonstrate a higher proportion of FGFR-overexpressing patients in their study than expected from The Cancer Genome Atlas (TCGA) data [9], e.g., 57% vs. 13% for HNSCC or 46% vs. 30% for squamous NSCLC, whereas data for gastric cancer (19% vs. 18%) or lung adenocarcinoma (11% vs. 12%) matched the database prediction. Interestingly and unexpectedly, it could also be demonstrated in this study that unusual alterations could be detected in the cohort of urothelial cancer patients. Here, 5.5% of enrolled patients showed overexpression of FGFR1 and several cases showed double positivity for FGFR1/2, FGFR1/3 or FGFR2/3 overexpression, which would not be detected by a FISH approach [112]. Still, a direct comparison of the prevalence of FGFR pathway alterations between different studies or databases needs to be conducted with some precautions, since different cut-offs or methods were applied that may lead to different results.
Taken together, these findings point to the direction that cancer patients with FGFR tumor overexpression, even in the absence of an underlying FGFR DNA alteration, could benefit from FGFR inhibitor therapy (Figure 2 & Table 2). As receptor tyrosine kinases, including FGFRs, are challenging targets for immunohistochemistry (IHC)-suited antibody generation, we discuss, in the following chapter, alternative ways to quantify FGFR expression using archival tumor biopsy specimens in order to identify patients most likely to benefit from FGFR inhibitor therapy.

Figure 2. Prevalence of FGFR alterations in selected tumor types. For each tumor type, the prevalence of amplifications, mutations, fusions or translocations and overexpression is highlighted according to [34–40,42–51,113–126]. Overexpression relates to protein overexpression as (usually) detected via immunohistochemistry. FGFR1 data marked with * for cholangiocarcinoma represent mRNA expression data. The most prevalent alteration is depicted in bold for each tumor type.

4.1. Immunohistochemistry

FGFR inhibitors currently available for clinical use are usually small-molecule inhibitors of the tyrosine kinase function of FGFRs. Therefore, directly detecting the drug target by immunohistochemistry was considered to be the best predictive and patient selection biomarker for these compounds and the technology would be readily available for decentralized testing in local pathology labs. Except for FGFR2-specific antibodies or FGFR2-targeting antibody drug conjugates, none of the small-molecule inhibitors listed in Table 1 currently use IHC as a predictive biomarker. The FGFR4-specific inhibitor Fisogatinib uses IHC to detect the FGFR ligand FGF19 [32], although recently, mRNA analysis was also used in this setting [127].
| Technology | Pros | Cons | Patient Population * | Prevalence |
|------------|------|------|----------------------|------------|
| **FGFR protein expression** | | | | |
| Immunohistochemistry | Broadly available, direct measure of receptor expression, keeps spatial resolution, short TAT | No single antibody, needs multiplexing for pan-FGFR inhibitors, Requires pathologist training or central testing | FGFR2b + gastric cancer | 30% |
| **FGFR mRNA expression** | | | | |
| PCR | Sensitive, cheap, short TAT, Easy to establish for each FGFR subtype | No preservation of spatial resolution | FGFR4 + HCC pts (Roblitinib) | Unknown |
| Nanostring | Sensitive, highly multiplex testing | Expensive, tumor content needs to be retrospectively calculated | FGFR1/2/3 + all comers (Rogaratinib) | Up to 25% |
| RNA-ISH | Sensitive, keeps spatial resolution, IHC-like workflow, short TAT, multiplex possible | Requires pathologist training or central testing | FGFR1/2/3 + all comers (AZD4547) | Unknown |
| RNAseq | Sensitive, highly multiplex testing | Expensive, long TAT (several weeks), no preservation of spatial resolution, | FGFR1/2/3 + all comers (Rogaratinib) | 25% |
| **FGFR DNA alterations** | | | | |
| FISH | Keeps spatial resolution | Requires fluorescence microscopy, multiplex possible | FGFR2 + gastric cancer (AZD4547) | 4–7% [11] |
| PCR | Short TAT (7 days) | No preservation of spatial resolution | FGFR2&3 fusion and FGFR3 mutations in urothelial carcinoma (QIAGEN’s FDA approved CDx therascreen® FGFR kit for Erdafitinib) | 20% [7] |
| NGS | Highly multiplex testing | Expensive, long TAT, no preservation of spatial resolution | FGFR2 fusion-positive iCCA (Foundation One™ as FDA approved CDx for Pemigatinib & Infigratinib) | 10% [91,98] |
| **FGF ligand** | | | | |
| IHC | Broadly available, direct measure of receptor expression, keeps spatial resolution, short TAT | No single antibody, needs multiplexing for pan-FGFR inhibitors, Requires pathologist training or central testing | FGF-19 serum levels in HCC (Fisogatinib) | 27% [32] |

CDx: Companion diagnostics; FDA: Food and Drug Administration; FISH: fluorescence in situ hybridization; HCC: hepatocellular carcinoma; iCCA: intrahepatic cholangiocellular carcinoma; IHC: immunohistochemistry; NGS: next generation sequencing; PCR: polymerase chain reaction; RNA-ISH: RNA in situ hybridization; RNA-seq: RNA sequencing; TAT: turnaround time. * Only patient populations that have been enrolled into FGFR inhibitor trials.
The use of immunohistochemistry for small-molecule FGFR inhibitors is further limited by the fact that these drugs are usually pan-FGFR inhibitors and, currently, there is no antibody available that detects all necessary isoforms simultaneously and with the needed sensitivity and specificity. Therefore, multiplex approaches using several isoform-specific antibodies would be needed, which are technically challenging to develop due to limited separability of chromogenic substrates. Fluorescence labels might overcome this technical limitation but would require specific technologies in the analysis labs, which limits the market access for this approach.

Overall, immunohistochemistry is not recommended as a predictive or patient selection biomarker for FGFR inhibitors.

4.2. FISH/CISH to Detect Gene Copy Number Variations

Several FGFR inhibitors have used fluorescence in situ hybridization (FISH) or chromogenic in situ hybridization (CISH) to detect gene copy number variations (CNV), since retrospective data showed a good correlation between high protein expression, as detected by immunohistochemistry, and CNV, e.g., for FGFR2 in gastric cancer [116], where a copy number gain is also associated with lymphatic invasion and poor prognosis [128]. Interestingly, the reverse correlation between CNV and protein expression could not be confirmed for FGFR1 in NSCLC patients, although FGFR1 amplification was also associated with poorer overall and disease-free survival here [129]. It is now confirmed that only high CNVs actually translate into high protein expression, which decreases the prevalence of FGFR-positive patients, usually to a low percentage of the overall population (e.g., 4% for FGFR2-amplified gastric cancer [130,131]). This raises the question for the right cut-off to achieve clinical efficacy, since only highly amplified cancers seem to be dependent on FGFR signaling and show higher sensitivity towards small-molecule inhibitors, such as AZD4547 [132]. Furthermore, there is great intratumor heterogeneity in FGFR CNVs and amplifications are not evenly distributed, which leads to the development of complex evaluation scores. In lung cancer, Schildhaus et al. proposed that high-level amplification is defined as FGFR1/centromere 8 (CEN8) ratio ≥ 2.0, or average number of FGFR1 signals per tumor cell nucleus ≥ 6 or the percentage of tumor cells containing ≥ 15 FGFR1 signals or large clusters ≥ 10% [133]. Such scores require extensive training of the pathologist to minimize interobserver variability, since selecting different scoring criteria may lead to different results. Interestingly, although FGF19 CNV could be detected by FISH and has been shown to correlate with response to the multi-kinase inhibitor sorafenib in HCC [134], current FGFR inhibitor trials either employ mRNA expression or protein overexpression via immunohistochemistry for FGF19 [32]. Similar to fluorescence-labeled immunohistochemistry, FISH requires specialized technical equipment that limits the broad application of this assay. CISH, which could overcome these limitations, is currently not used in any clinical trial.

4.3. mRNA Expression Technologies

RNA in situ hybridization (RNA-ISH) was considered to be challenging in clinical trial settings. However, the introduction of the RNAscope technology in 2012 allows for sensitive and specific detection of individual mRNA molecules, also in paraffin-embedded tissue specimens, due to a novel probe design. It preserves spatial resolution and tissue architecture and can be used with chromogenic or fluorescent detection systems and is, therefore, highly multiplexable and results are well quantifiable [135].

The Nanostring nCounter technology was first introduced in 2008. It is a highly multiplexable direct measurement of mRNA expression with high sensitivity and reproducibility and low detection limit, also from paraffin-embedded tissues [136]. In contrast to mRNA in situ hybridization, but similar to PCR or sequencing technologies, it does not maintain the spatial resolution of signals, since a direct digital readout is performed. However, new algorithms allow one to recalculate stroma/tumor content and, thus, provide additional information on signal distribution.
Preclinical data indicated that, actually, mRNA expression is a better predictor for FGFR sensitivity than CNV or other biomarkers [103,106]. Schuler et al. were the first to adopt those technologies for prospective selection of patients in a Phase 1 study in an indication-agnostic manner [9]. In addition to demonstrating the technical feasibility and robustness in a global multi-center trial, they could demonstrate that mRNA technologies identified a much broader patient population (including patients without apparent genetic alteration) than anticipated, thus, broadening the potential to bring benefit to patients. These data were recently confirmed for FGFR1–4 mRNA expression in breast cancer [137].

While Nanostring technology is highly sensitive and multiplexable, it is also considered expensive and requires bioinformatics workup to calculate tumor content. RNAscope offers the advantage of maintaining the spatial resolution, which may provide useful additional information on distribution of signals. It is considered fast and provides an IHC-like readout but does require intensive training of the pathologist or a central reading in clinical studies to minimize interobserver variability.

In addition, several PCR-based technologies (e.g., digital droplet PCR or quantitative reverse transcriptase PCR) have been developed and used in clinical trials, usually only on a retrospective basis and not for patient selection. This is also the case for RNA sequencing.

4.4. Next-Generation DNA Sequencing (NGS)

Massive parallel sequencing, now usually called next-generation sequencing (NGS), was developed in the mid-1990s and allows for the rapid reading of DNA fragments up to 400 base pairs in length and a maximum readout of up to 1 terabase per run. This provides information on multiple genes or genetic aberrations, including mutations, fusions and copy number variations, which is of special interest for FGFR inhibitors. First kits, e.g., Foundation One, have received FDA approval as a CDx test for various drugs and indications. Currently, pemigatinib and infgratinib use Foundation One as an FDA-approved companion diagnostics test (CDx) for FGFR2 fusions in intrahepatic cholangiocellular carcinoma [138].

In general, NGS approaches are considered the gold standard for molecular testing in cancer. Their availability and reimbursement are constantly growing, since they allow one to obtain information on numerous druggable genetic alterations at once, thus, limiting the need for tissue and specific sequencing requests [139]. NGS results are complex and contain information in several genetic alterations, which requires expert discussion in molecular tumor boards, since, currently, no hierarchy of results is established, which would guide treatment decisions in the case of multiple druggable hits. The benefit of obtaining multiple insights at once is, of course, that guidance on resistance mechanisms and on sequencing of therapeutic approaches could be discussed up front.

A limitation of NGS is its rather long turnaround time of up to 4 weeks and the comparably high costs of the approach. The latter one is a major hurdle when using NGS to identify rare genetic alterations with low prevalence in a certain population, which often makes the use of NGS prohibitive, even in clinical trials.

4.5. NGS of ctDNA (Liquid Biopsies)

To overcome limitations related to tissue-based testing, such as invasiveness, biopsy sampling error, intra-tumor heterogeneity or scarcity of available tissues, liquid biopsies have been included in various clinical trials. Here, circulating free tumor DNA (ctDNA) can be analyzed by means of next-generation sequencing and alterations, meaning mutations or fusions can easily be detected. Furthermore, the level of ctDNA itself has been shown to be an early prognostic marker correlated to disease recurrence in, e.g., NSCLC or colorectal cancer [140-142].

Jogo et al. demonstrated that ctDNA analysis detects FGFR2 amplification in gastric cancer at a higher frequency than tissue analysis (7.7% vs. 2.6–4.4%). They could also identify patients where FGFR2 amplification was detectable only in liquid biopsy but not in a paired tissue sample and that these cases had an overall worse prognosis [110]. Further
data are needed to validate if only high-level amplifications (CNV > 5) translate into ctDNA positivity or if also lower-copy-number changes could be detected in liquid biopsies [132].

An important application of NGS analyses to ctDNA samples is the monitoring of resistance development. It was demonstrated that FGFR inhibitor treatment can lead to secondary mutations in FGFR2 cholangiocarcinomas with FGFR2 alterations, which drives resistance. Here, ctDNA analyses were applied longitudinally and could overcome the observed intratumor heterogeneity and polyclonality in assessing resistance mutations [143,144]. This approach was also used to inform treatment decisions in patients who developed resistance against infigratinib or Debio 1347 and who could still benefit from subsequent treatment with the irreversible FGFR inhibitor futibatinib [145]. Interestingly, resistance to the CDK4/6 inhibitor ribociclib, in combination with fulvestrant in ER+ breast cancer, can be mediated by amplification of FGFR1. ctDNA analysis from the registrational MONALEESA-2 trial confirmed that patients with FGFR1 amplification had shorter progression-free survival than wild-type patients [146].

Overall, these data demonstrate the high diagnostic, prognostic and predictive value of NGS-based detection of FGFR pathway alterations. Specifically, the sophisticated longitudinal monitoring of resistance development has great potential to improve treatment strategies due to advanced and adaptive therapeutic schedules for patients.

5. Discussion

The low concordance between FGFR amplification and FGFR overexpression, as described for large datasets from NSCLC [104] and HNSCC [106] patients, questions to what extent the pre-selection of patients based on FGFR amplifications may have led to the failure of early clinical trials with FGFR inhibitors [147]. In addition, even for the very same FGFR alteration within the same tumor type, highly different prevalence data are reported across published data: a recent meta-analysis on the frequency of FGFR1 gene amplification in NSCLC evaluating twenty-three studies (5252 patients) revealed a 10-fold difference in the prevalence, ranging from only 4.9 to up to 49% [148]. This high variability raises the question if an additional layer of complexity to define an FGFR-positive patient is based on the method used to detect the FGFR alteration, which all have a different sensitivity and/or specificity. In addition, the definition of FGFR positivity might be improved by applying, in parallel, two different methods to detect FGFR alterations or to perform even orthogonal assays, e.g., evaluating FGFR DNA alterations and FGFR expression levels using the same tumor tissue biopsy specimen. To date, only limited data on FGFR positivity, confirmed by two different readouts, are available for patients treated with an FGFR inhibitor. However, recent data from the Phase 2/3 study FORT-1, evaluating rogaratinib in first-line urothelial cancer, revealed a very high response rate of 52.4% in a small subgroup of patients pre-selected for FGFR1/3 mRNA overexpression and retrospectively confirmed of having, in addition, either an underlying FGFR3-activating mutation or an FGFR3 gene fusion compared to the ORR of 19.5% in patients being positive for FGFR1/3 mRNA overexpression, regardless of whether an underlying FGFR DNA alteration was detected [149]. This points to a higher benefit from FGFR inhibitor therapy in patients with FGFR positivity, confirmed by two different readouts and, ultimately, leads to the provocative question of whether the therapeutic impact of existing FGFR inhibitors is rather limited by the difficulties to define an FGFR-positive tumor than by the drugs themselves.

In addition to technical reasons for choosing a certain predictive biomarker assay, also, clinical and molecular factors need to be taken into consideration to find the best therapy for patients (Figure 3). Still, several key questions remain elusive when selecting a predictive FGFR biomarker assay.
which leads to a basic physiologic signaling to maintain metabolic and tissue homeosta-
sis. Thus, the fact that non-malignantly transformed somatic cells
are active in normal body cells [16,64] could also explain the early and rapid development
of resistance bypass pathways under FGFR inhibitor treatment, as demonstrated for, e.g.,
the upregulation of ErbB family members after infgratinib treatment or for upregulation
of the EGFR pathway [156,157]. A common downstream mediator of receptor tyrosine
kinases is the PI3K/AKT/MAPK pathway. Alterations in this pathway, downstream of
the receptor, have also been described to confer resistance to FGFR inhibitors [158,159] and
could, in turn, be overcome again by combination therapy with an MEK inhibitor [160].
Insights into the parallel occurrence of such resistance mutations could, therefore, improve

Figure 3. Factors influencing the selection of a predictive biomarker assay for FGFR-inhibitor
therapies. Alterations in FGFR1-4 impact on the predictivity of a biomarker assay. In addition to
the molecular biology of the alterations (CNV, fusion, mutation, etc.), also, the underlying tumor
tility (histology), the clinical staging (e.g., muscle-invasive vs. non-muscle-invasive bladder cancer),
tissue availability and the assay technology with different target readouts (protein, mRNA, DNA)
determine which FGFR targeting therapy would bring benefit to a patient.

First, gene fusions are considered strong oncogenic drivers and recent clinical data
show strong efficacy of compounds targeting, e.g., neuregulin 1 (NRG1) [150–152], neu-
rotrophic tyrosine kinase receptor (NTRK) [152,153] or anaplastic lymphoma kinase
(ALK) [154,155] fusions in patients across various tumor types. Despite selecting patients
for FGFR gene fusions, clinical responses to FGFR inhibitors seem less deep and less
pronounced than compared to other fusion-specific agents, such as Larotrectinib. This
may be due to the complex crosstalk and redundancy within the FGFR signaling network,
which leads to a basic physiologic signaling to maintain metabolic and tissue homeo-
stasis functions [16,64]. For example, whilst the normal, non-fusion-bearing NTRK protein
does not play any role outside the central nervous system in adults and the physiological
roles of ALK1 proteins in adults are still a matter of debate, FGFR proteins exert many
physiological functions. Thus, the fact that non-malignantly transformed somatic cells
express baseline FGFRs (e.g., FGFR3 in normal urothelial cells) separates FGFR proteins
clearly from NTRK proteins, where adult somatic cells lack expression and, thus, down-
stream signaling. In addition, the continuous physiological FGFR background signaling
active in normal body cells [16,64] could also explain the early and rapid development
of resistance bypass pathways under FGFR inhibitor treatment, as demonstrated for, e.g.,
the upregulation of ErbB family members after infgratinib treatment or for upregulation
of the EGFR pathway [156,157]. A common downstream mediator of receptor tyrosine
kinases is the PI3K/AKT/MAPK pathway. Alterations in this pathway, downstream of
the receptor, have also been described to confer resistance to FGFR inhibitors [158,159] and
could, in turn, be overcome again by combination therapy with an MEK inhibitor [160].
Insights into the parallel occurrence of such resistance mutations could, therefore, improve
the response rates of FGFR inhibitors and favor the use of NGS panel approaches to select patients for FGFRi treatment. Interestingly, also for the approved FGFR inhibitors, differences in efficacy were observed based on the underlying FGFR DNA alteration. For pemigatinib, all observed responses were limited to FGFR2 fusion-positive cholangiocellular carcinoma and no confirmed responses were seen in other FGFR alterations [98]. In urothelial cancer, in contrast, long-term follow-up of a phase 2 study of erdafitinib revealed that duration of response and overall survival were generally similar between patients with FGFR mutations and those with FGFR fusions [161], which renders clinical decision-making challenging. In comparison to other growth factor receptor pathway inhibitors, e.g., Osimertinib against EGFR, agents targeting FGFRs seem have a lower overall potency, which indicates that tumor cells may be less addicted to complex FGFR signaling pathways with their multiple redundancies.

Second, it is intriguing that the same FGFR DNA alteration (mutation, fusion, amplification) leads to different sensitivity to FGFR inhibitors, depending on the underlying histologic subtype. Across several compounds, FGFR2 fusions were most sensitive in intrahepatic cholangiocellular carcinoma (ihCC) and FGFR3 mutations showed the best responses in urothelial cancers. FGFR2 amplification gave positive results in gastric cancer but disappointed in, e.g., breast cancer or NSCLC. It is unclear why different tumor types show different dependencies on these FGFR pathway alterations, even when DNA-independent biomarkers, such as mRNA overexpression, are applied. So far, only limited data are available on potential co-mutations or further downstream alterations, but it is obvious that tumor type and histology matter [74].

Third, the overall prognostic and predictive value of (different) FGFR alterations remains unclear. It is also unclear what a biologically meaningful cut-off for the different assay formats discussed above would be and a clear threshold for FGFR positivity is currently not available. As an example, approved CDx tests for FGFR inhibitor treatment, based on FGFR mutations or fusions, apply a cut-off for FGFR positivity as a mutant allele fraction (MAF) of at least 5%, whereas an NGS-based mutation test may easily detect an MAF of 0.1% or even less. However, in contrast to the established cut-off for the MAF for EGFR-activating mutations shown to be clinically meaningful for EGFR inhibitor treatment [162], no such correlation of MAF and clinical response has been shown for any FGFR inhibitor to date. The same applies for the correlation between the degree of FGFR protein or FGFR mRNA overexpression and clinical response when being used for patient selection in clinical trials or for the cut-off definition of a clinically meaningful copy number gain.

Only limited data are available that confirm a negative prognosis for urothelial cancer patients with FGFR genomic alterations [163], while other data could not establish a correlation to overall survival (OS) or progression-free survival (PFS) or response to systemic therapy [164]. Several studies recently investigated the combination of FGFR inhibitors with immune checkpoint inhibitors. While a recent study from Denmark could not identify a statistically significant correlation between FGFR3 amplifications or mutations to PD-L1 expression in primary urothelial carcinomas [58], Sweis et al. showed that an activated FGFR3 pathway is linked to non-T-cell-inflamed tumors, which are characterized by poorer prognosis and resistance to immune checkpoint inhibitors [56]. This may be due to the activation of neural-precursor-cell-expressed developmentally down-regulated protein 4 (NEDD4), an E3 ubiquitin ligase, by activated FGFR3 that could lead to proteasomal degradation of PD-L1, indicating the potential of combination therapy in this setting [165]. FGFR3 mutations, therefore, seem to be a negative predictor to immunotherapy response in urothelial cancer. However, promising data from erdafitinib and rogaratinib combination trials with PD-1 antibodies indicate that parallel inhibition of overactive FGFR signaling in urothelial cancer may be a pre-requisite to sensitize tumors to the benefit of subsequent checkpoint inhibitor therapy [166,167].
6. Conclusions

Drugs targeting the FGFR pathway have matured and received approval for various cancer indications, which require prospective biomarker testing. Several technologies, ranging from immunohistochemistry to PCR or sequencing technologies and to gene expression approaches, have been investigated in clinical trials and provide different advantages and limitations. Next-generation sequencing is approved as a companion diagnostic kit for two compounds but further understanding of the role of distinct alterations (e.g., gene fusions vs. mutations vs. overexpression) is urgently needed, as a one-fits-all approach does not seem successful for FGFR inhibitors. The available clinical data indicate that the nature of the alteration and the underlying cancer disease itself significantly impact the predictivity of those biomarkers and more research is needed to obtain clarity for clinicians and patients on what biomarker cut-off and what test achieve the best results in a certain disease context.

Regarding the knowledge of advantages and limitations in FGFR tests, the following recommendations can be given: In situ hybridization approaches allow one to determine the gene copy number, which seems to be a less reliable predictor of treatment outcome. Immunohistochemistry is currently not recommended as a screening method in view of the possibility of NGS assays. Depending on the available patient sample (including the possibility to achieve tumor content enrichment through tissue microdissection or the alternative use of liquid biopsies), molecular pathological testing of potential FGFR mutations or fusions should be performed using DNA/RNA-based NGS platforms.

Author Contributions: D.N., P.E. and M.O. carried out conceptualization. M.O., S.S. and D.N. performed literature research. M.O., D.N., P.E., H.N. and S.S. wrote the manuscript. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Conflicts of Interest: D.N. received honoraria for advisory function at Boehringer Ingelheim and Lilly. S.S. received honoraria and financial support from Amgen, AstraZeneca, Bayer, BMS, Esai, Lilly, Merck KGaA, MSD, Pierre-Fabre, Roche, Sanofi, Sevier, Taiho and Takeda. The funders had no role in the design of this manuscript, in the collection, analyses, or interpretation of data; in the writing of the manuscript, or in the decision to publish the results. All other authors declare no conflict of interest related to this manuscript.

References
1. Hanahan, D. Hallmarks of Cancer: New Dimensions. Cancer Discov. 2022, 12, 31–46. [CrossRef] [PubMed]
2. Weiss, F.; Lauffenburger, D.; Friedl, P. Towards Targeting of Shared Mechanisms of Cancer Metastasis and Therapy Resistance. Nat. Rev. Cancer 2022, 22, 157–173. [CrossRef] [PubMed]
3. Liu, L.; Wang, C.; Li, S.; Bai, H.; Wang, J. Tumor Immune Microenvironment in Epidermal Growth Factor Receptor-Mutated Non-Small Cell Lung Cancer before and after Epidermal Growth Factor Receptor Tyrosine Kinase Inhibitor Treatment: A Narrative Review. Transl. Lung Cancer Res. 2021, 10, 3823–3839. [CrossRef]
4. National Guideline Alliance (UK). Use of Molecular Biomarkers to Guide Systemic Therapy: Colorectal Cancer (Update): Evidence Review B1; NICE Evidence Reviews Collection; National Institute for Health and Care Excellence (NICE): London, UK, 2020; ISBN 978-1-4731-3657-1.
5. Jørgensen, J.T.; Winther, H.; Askaa, J.; Andresen, L.; Olsen, D.; Mollerup, J. A Companion Diagnostic With Significant Clinical Impact in Treatment of Breast and Gastric Cancer. Front. Oncol. 2021, 11, 676939. [CrossRef] [PubMed]
6. Papachristos, A.; Sivolapenko, G.B. Pharmacogenomics, Pharmacokinetics and Circulating Proteins as Biomarkers for Bevacizumab Treatment Optimization in Patients with Cancer: A Review. J. Pers. Med. 2020, 10, 79. [CrossRef] [PubMed]
7. Loriot, Y.; Necchi, A.; Park, S.H.; Garcia-Donas, J.; Huddart, R.; Burgess, E.; Fleming, M.; Rezazadeh, A.; Mellard, B.; Varlamov, S.; et al. Erdafitinib in Locally Advanced or Metastatic Urothelial Carcinoma. N. Engl. J. Med. 2019, 381, 338–348. [CrossRef]
8. Kang, C. Infigratinib: First Approval. Drugs 2021, 81, 1355–1360. [CrossRef]
9. Schuler, M.; Cho, B.C.; Sayehli, C.M.; Navarro, A.; Soo, R.A.; Richly, H.; Cassier, P.A.; Tai, D.; Penel, N.; Nogova, L.; et al. Rogaratinib in Patients with Advanced Cancers Selected by FGFR mRNA Expression: A Phase 1 Dose-Escalation and Dose-Expansion Study. *Lancet Oncol.* **2019**, *20*, 1454–1466. [CrossRef]

10. Subbiah, V.; Iannotti, N.O.; Gutierrez, M.; Smith, D.C.; Feliz, L.; Lihou, C.F.; Tian, C.; Silverman, I.M.; Ji, T.; Saleh, M. FIGHT-101, a First-in-Human Study of Potent and Selective FGFR 1-3 Inhibitor Pemigatinib in Pan-Cancer Patients with FGF/FGFR Alterations and Advanced Malignancies. *Ann. Oncol.* **2022**, *33*, 522–533. [CrossRef]

11. Van Cutsem, E.; Bang, Y-J.; Mansoor, W.; Petty, R.D.; Chao, Y.; Cunningham, D.; Ferry, D.R.; Smith, N.R.; Frewer, P.; Ratnayake, J.; et al. A Randomized, Open-Label Study of the Efficacy and Safety of AZD4547 Monotherapy versus Paclitaxel for the Treatment of Advanced Gastric Adenocarcinoma with FGFR2 Polysomy or Gene Amplification. *Ann. Oncol.* **2017**, *28*, 1316–1324. [CrossRef]

12. Turner, N.; Gros, R. Fibroblast Growth Factor Signalling: From Development to Cancer. *Nat. Rev. Cancer* **2010**, *10*, 116–129. [CrossRef]

13. Kelleher, F.C.; O’Sullivan, H.; Smyth, E.; McDermott, R.; Viterbo, A. Fibroblast Growth Factor Receptors, Developmental Corruption and Malignant Disease. *Carcinogenesis* **2013**, *34*, 2198–2205. [CrossRef] [PubMed]

14. Eswarakumar, V.P.; Lax, I.; Schlessinger, J. Cellular Signaling by Fibroblast Growth Factor Receptors. *Cytokine Growth Factor Rev.* **2005**, *16*, 139–149. [CrossRef] [PubMed]

15. Zhang, X.; Ibrahim, O.A.; Olsen, S.K.; Umemori, H.; Mohammadi, M.; Ornitz, D.M. Receptor Specificity of the Fibroblast Growth Factor Family. The Complete Mammalian FGF Family. *J. Biol. Chem.* **2006**, *281*, 15694–15700. [CrossRef] [PubMed]

16. Heroult, M.; Ellinghaus, P.; Ince, S.; Ocker, M. Fibroblast Growth Factor Receptor Signaling in Cancer Biology and Treatment. *Curr. Signal Transduct. Ther.* **2014**, *9*, 15–25. [CrossRef]

17. Ong, S.H.; Guy, G.R.; Hadari, Y.R.; Laks, S.; Gotoh, N.; Schlessinger, J.; Lax, I. FRS2 Proteins Recruit Intracellular Signaling Pathways by Diverse Targets on Fibroblast Growth Factor and Nerve Growth Factor Receptors. *Mol. Cell. Biol.* **2000**, *20*, 979–989. [CrossRef]

18. Ong, S.H.; Hadari, Y.R.; Gotoh, N.; Guy, G.R.; Schlessinger, J.; Lax, I. Stimulation of Phosphatidylinositol 3-Kinase by Fibroblast Growth Factor Receptor Is Mediated by Coordinated Recruitment of Multiple Docking Proteins. *Proc. Natl. Acad. Sci. USA* **2001**, *98*, 6074–6079. [CrossRef]

19. Kouhara, H.; Hadari, Y.R.; Spivak-Kroizman, T.; Schilling, J.; Bar-Sagi, D.; Lax, I.; Schlessinger, J. A Lipid-Anchored Grb2-Binding Protein That Links FGF-Receptor Activation to the Ras/ MAPK Signaling Pathway. *Cell* **1997**, *89*, 693–702. [CrossRef]

20. Hart, K.C.; Robertson, S.C.; Smith, D.C.; Feliz, L.; Lihou, C.F.; Tian, C.; Silverman, I.M.; Ji, T.; Saleh, M. FIGHT-101, a First-in-Human Study of Potent and Selective FGFR 1-3 Inhibitor Pemigatinib in Pan-Cancer Patients withFGF/FGFR Alterations and Advanced Malignancies. *Ann. Oncol.* **2022**, *33*, 522–533. [CrossRef]

21. Quintanal-Villalonga, C.; Ojeda, S.; Zugazagoitia, J.; Muñoz-Galván, S.; Lopez-Rios, F.; et al. FGFR1 and FGFR4 Oncogenicity Depends on N-Cadherin and Their Co-Expression May Predict FGFR-Targeted Therapy Efficacy. *ElBioMedicine* **2020**, *53*, 102683. [CrossRef]

22. Nguyen, T.; Duchesne, L.; Sankara Narayana, G.H.N.; Bobgetto, N.; Fernig, D.D.; Uttamrao Murade, C.; Ladoux, B.; Mège, R.-M. Enhanced Cell-Cell Contact Stability and Decreased N-Cadherin-Mediated Migration upon Fibroblast Growth Factor Receptor-N-Cadherin Cross Talk. *Oncogene* **2019**, *38*, 6283–6300. [CrossRef] [PubMed]

23. Nguyen, T.; Mége, R.M. N-Cadherin and Fibroblast Growth Factor Receptors Crosstalk in the Control of Developmental and Cancer Cell Migrations. *Eur. J. Cell Biol.* **2016**, *95*, 415–426. [CrossRef]

24. Buchtova, M.; Oralova, V.; Aklian, A.; Masek, J.; Vesela, I.; Ouyang, Z.; Obadalova, T.; Konecna, Z.; Spoustaova, T.; Pospisilova, T.; et al. Fibroblast Growth Factor and Canonical WNT/β-Catenin Signaling Cooperate in Suppression of Chondrocyte Differentiation in Experimental Models of FGFR and Canonical WNT Signaling in Cartilage. *Biochim. Biophys. Acta* **2015**, *1852*, 839–850. [CrossRef]

25. Mavila, N.; James, D.; Utley, S.; Cui, N.; Coblens, O.; Mak, K.; Rountree, C.B.; Kahn, M.; Wang, K.S. Fibroblast Growth Factor Receptor-Mediated Activation of AKT-β-Catenin-CBP Pathway Regulates Survival and Proliferation of Murine Hepatoblasts and Hepatic Tumor Initiating Stem Cells. *PLoS ONE* **2012**, *7*, e50401. [CrossRef]

26. Carstens, J.M.; Shahi, P.; Van Tsang, S.; Smith, B.; Creighton, C.J.; Zhang, Y.; Seamans, A.; Seethammagari, M.; Vendula, I.; Levitt, J.M.; et al. FGFR1-WNT-TGF-β Signaling in Prostate Cancer Mouse Models Recapitulates Human Reactive Stroma. *Cancer Res.* **2014**, *74*, 609–620. [CrossRef] [PubMed]

27. Jaidée, R.; Kukongviriyapan, V.; Senggunprai, L.; Prawan, A.; Jusakul, A.; Laphanuwat, P.; Kongpetch, S. Inhibition of FGFR2 Enhances Chemosensitivity to Gemcitabine in Cholangiocarcinoma through the AKT/MTOR and EMT Signaling Pathways. *Life Sci.* **2022**, *296*, 120427. [CrossRef]

28. Roy Burman, D.; Das, S.; Das, C.; Bhattacharya, R. Alternative Splicing Modulates Cancer Aggressiveness: Role in EMT/Metastasis and Chemoresistance. *Mol. Biol. Rep.* **2021**, *48*, 897–914. [CrossRef] [PubMed]

29. Miki, T.; Bottaro, D.P.; Fleming, T.P.; Smith, C.L.; Chan, A.M.; Aaronson, S.A. Determination of Ligand-Binding Specificity by Alternative Splicing: Two Distinct Growth Factor Receptors Encoded by a Single Gene. *Proc. Natl. Acad. Sci. USA* **1992**, *89*, 246–250. [CrossRef]

30. Wescle, J.; Haglund, K.; Haugsten, E.M. Fibroblast Growth Factors and Their Receptors in Cancer. *Biochem. J.* **2011**, 437, 199–213. [CrossRef]

31. Liu, H.; Niu, D.; Tham Sjin, R.T.; Dubrovskiy, A.; Zhu, Z.; McDonald, J.J.; Fahnee, K.; Wang, Z.; Munson, M.; Scholte, A.; et al. Discovery of Selective, Covalent FGFR4 Inhibitors with Antitumor Activity in Models of Hepatocellular Carcinoma. *ACS Med. Chem. Lett.* **2020**, *11*, 1899–1904. [CrossRef]
32. Kim, R.D.; Sarker, D.; Meyer, T.; Yau, T.; Macarulla, T.; Park, J.-W.; Choo, S.P.; Hollebecque, A.; Sung, M.W.; Lim, H.-Y.; et al. First-in-Human Phase I Study of Fisogatinib (BLU-554) Validates Ablative FGFR19 Signaling as a Driver Event in Hepatocellular Carcinoma. *Cancer Discov.* 2019, 9, 1696–1707. [CrossRef] [PubMed]

33. Zehir, A.; Benayed, R.; Shah, R.H.; Syed, A.; Middha, S.; Kim, H.R.; Srinivasan, P.; Gao, J.; Chakravarty, D.; Devlin, S.M.; et al. Mutational Landscape of Metastatic Cancer Revealed from Prospective Clinical Sequencing of 10,000 Patients. *Nat. Med.* 2017, 23, 703–713. [CrossRef] [PubMed]

34. Dutt, A.; Salvesen, H.B.; Chen, T.-H.; Ramos, A.H.; Onofrio, R.C.; Hatton, C.; Nicoletti, R.; Winckler, W.; Grewal, R.; Hanna, M.; et al. FGF/FGFR Signaling Pathway Involved Resistance in Various Cancer Types. *Clin. Epigenetics* 2016, 22, 259–267. [CrossRef]

35. Matsumoto, K.; Arao, T.; Hamaguchi, T.; Shimada, Y.; Kato, K.; Oda, I.; Taniguchi, H.; Koizumi, F.; Yanagihara, K.; Sasaki, H.; et al. Clinicopathologic Significance in Gastric Cancer. *Clin. Cancer Res.* 2010, 636–647. [CrossRef] [PubMed]

36. Zhou, Y.; Wu, C.; Lu, G.; Hu, Z.; Chen, Q.; Du, X. FGF/FGFR Signaling Pathway Involved Resistance in Various Cancer Types. *J. Cancer* 2020, 11, 2000–2007. [CrossRef] [PubMed]

37. Chang, J.; Liu, X.; Wang, S.; Zhang, Z.; Wu, Z.; Zhang, X.; Li, J. Prognostic Value of FGFR Gene Amplification in Patients with Different Types of Cancer: A Systematic Review and Meta-Analysis. *PLoS ONE* 2014, 9, e105524. [CrossRef]

38. Ahmad, I.; Iwata, T.; Leung, H.Y. Mechanisms of FGFR-Mediated Carcinogenesis. *Biochim. Biophys. Acta* 2012, 1823, 850–860. [CrossRef]

39. Reis-Filho, J.S.; Simpson, P.T.; Turner, N.C.; Lambros, M.B.; Jones, C.; Mackay, A.; Grigoriadis, A.; Sarrio, D.; Savage, K.; Dexter, T.; et al. Drug-Sensitive FGFR2 Mutations in Endometrial Carcinoma. *Proc. Natl. Acad. Sci. USA* 2008, 105, 8713–8717. [CrossRef]

40. Gust, K.M.; McConkey, D.J.; Awrey, S.; Hegarty, P.K.; Qing, J.; Bondaruk, J.; Ashkenazi, A.; Czerniak, B.; Dinney, C.P.; Black, P.C. Fibroblast Growth Factor Receptor 3 Is a Rational Therapeutic Target in Bladder Cancer. *Mol. Cancer Ther.* 2013, 12, 1245–1254. [CrossRef]

41. Repetto, M.; Crimini, E.; Giugliano, F.; Morganti, S.; Belli, C.; Curigliano, G. Selective FGFR/FGF Pathway Inhibitors: Inhibition Strategies, Clinical Activities, Resistance Mutations, and Future Directions. *Expert Rev. Clin. Pharmacol.* 2021, 14, 1233–1252. [CrossRef]

42. Weiss, J.; Sos, M.L.; Seidel, D.; Peifer, M.; Zander, T.; Heuckmann, J.M.; Ullrich, R.T.; Menon, R.; Maier, S.; Soltermann, A.; et al. Frequent and Focal FGFR1 Amplification Associates with Therapeutically Tractable FGFR1 Dependency in Squamous Cell Lung Cancer. *Sci. Transl. Med.* 2010, 2, 62ra93. [CrossRef] [PubMed]

43. Yang, W.; Yao, Y.-W.; Zeng, J.-L.; Liang, W.-J.; Wang, L.; Bai, C.-Q.; Liu, C.-H.; Song, Y. Prognostic Value of FGFR1 Gene Copy Number in Patients with Non-Small Cell Lung Cancer: A Meta-Analysis. *J. Thorac. Dis.* 2014, 6, 803–809. [CrossRef] [PubMed]

44. Mao, P.; Cohen, O.; Kowalski, K.J.; Kusiel, J.G.; Buendia-Buendia, J.E.; Cuoco, M.S.; Wander, S.A.; Waks, A.G.; Nayar, U.; et al. Acquired FGFR and FGF Alterations Confer Resistance to Estrogen Receptor (ER) Targeted Therapy in ER+ Metastatic Breast Cancer. *Clin. Cancer Res.* 2020, 26, 5974–5989. [CrossRef]

45. Reis-Filho, J.S.; Simpson, P.T.; Turner, N.C.; Lambros, M.B.; Jones, C.; Mackay, A.; Grigoriadis, A.; Sarrio, D.; Savage, K.; Dexter, T.; et al. FGFR1 Emerges as a Potential Therapeutic Target for Lobular Breast Carcinomas. *Clin. Cancer Res.* 2006, 12, 6652–6662. [CrossRef] [PubMed]

46. Bao, Y.; Gabrielpillai, J.; Dietrich, J.; Zarbl, R.; Strieth, S.; Schröck, F.; Dietrich, D. Fibroblast Growth Factor (FGF), FGF Receptor (FGFR), and Cyclin D1 (CCND1) DNA Methylation in Head and Neck Squamous Cell Carcinomas Is Associated with Transcriptional Activity, Gene Amplification, Human Papillomavirus (HPV) Status, and Sensitivity to Tyrosine Kinase Inhibitors. *Clin. Epigenetics* 2021, 13, 228. [CrossRef] [PubMed]

47. Zehir, A.; Benayed, R.; Shah, R.H.; Syed, A.; Middha, S.; Kim, H.R.; Srinivasan, P.; Gao, J.; Chakravarty, D.; Devlin, S.M.; et al. Identification of Targetable FGFR gene fusions in diverse cancers. *Cancer Discov.* 2013, 3, 636–647. [CrossRef]

48. Li, Y.; Qiu, X.; Wang, X.; Liu, H.; Geck, R.C.; Tewari, A.K.; Xiao, T.; Font-Tello, A.; Lim, K.; Jones, K.L.; et al. FGFR-Inhibitor-Mediated Dismissal of SWI/SNF Complexes from YAP-Dependent Enhancers Induces Adaptive Therapeutic Resistance. *Nat. Cell Biol.* 2021, 23, 1187–1198. [CrossRef] [PubMed]
54. Bogatyrova, O.; Mattsson, J.S.M.; Ross, E.M.; Sanderson, M.P.; Backman, M.; Botling, J.; Brunnström, H.; Kurppa, P.; La Fleur, L.; Strell, C.; et al. FGFR1 Overexpression in Non-Small Cell Lung Cancer Is Mediated by Genetic and Epigenetic Mechanisms and Is a Determinant of FGFR1 Inhibitor Response. Eur. J. Cancer 2021, 151, 136–149. [CrossRef] [PubMed]

55. Li, P.; Huang, T.; Zou, Q.; Liu, D.; Wang, Y.; Tan, X.; Wei, Y.; Qiu, H. FGFR2 Promotes Expression of PD-L1 in Colorectal Cancer via the JAK/STAT3 Signaling Pathway. J. Immunol. 2019, 202, 3065–3075. [CrossRef] [PubMed]

56. Siewe, R.F.; Spranger, S.; Bao, R.; Paner, G.P.; Stadler, W.M.; Steinberg, G.; Gajewski, T.F. Molecular Drivers of the Non-T-Cell-Inflamed Tumor Microenvironment in Urothelial Bladder Cancer. Cancer Immunol. Res. 2016, 4, 563–568. [CrossRef] [PubMed]

57. Rose, T.L.; Weir, W.H.; Mayhew, G.M.; Shibata, Y.; Eulitt, P.; Uronis, J.M.; Zhou, M.; Nielsen, M.; Smith, A.B.; Woods, M.; et al. Fibroblast Growth Factor Receptor 3 Alterations and Response to Immune Checkpoint Inhibition in Metastatic Urothelial Cancer: A Real World Experience. Br. J. Cancer 2021, 125, 1251–1260. [CrossRef]

58. Grantzau, T.; Toft, B.G.; Melchior, L.C.; Elversang, J.; Stormoen, D.B.; Orland, L.H.; Pappot, H. PD-L1 Expression and FGFR-Mutations among Danish Patients Diagnosed with Metastatic Urothelial Carcinoma: A Retrospective and Descriptive Study. APMIS Acta Pathol. Microbiol. Immunol. Scand. 2022, 130, 498–506. [CrossRef]

59. Akhand, S.S.; Liu, Z.; Purdy, S.C.; Abdullah, A.; Lin, H.; Cresswell, G.M.; Ratliff, T.L.; Wendt, M. Pharmacologic Inhibition of FGFR Modulates the Metastatic Tumor Microenvironment and Promotes Response to Immune Checkpoint Blockade. Cancer Immunol. Res. 2020, 8, 1542–1553. [CrossRef]

60. Deng, H.; Kan, A.; Lyu, N.; Mu, L.; Han, Y.; Liu, L.; Zhang, Y.; Duan, Y.; Liao, S.; Li, S.; et al. Dual Vascular Endothelial Growth Factor Receptor and Fibroblast Growth Factor Receptor Inhibition Elicits Antitumor Immunity and Enhances Programmed Cell Death-1 Checkpoint Blockade in Hepatocellular Carcinoma. Liver Cancer 2020, 9, 338–357. [CrossRef]

61. Palakurthi, S.; Kuraguchi, M.; Zacharek, S.J.; Zudaire, E.; Huang, W.; Bonal, D.M.; Liu, J.; Dhaneswar, A.; DePeaux, K.; Gowaski, M.R.; et al. The Combined Effect of FGFR Inhibition and PD-1 Blockade Promotes Tumor-Intrinsic Induction of Antitumor Immunity. Cancer Immunol. Res. 2019, 7, 1457–1471. [CrossRef]

62. Wu, Y.; Yi, Z.; Li, J.; Wei, Y.; Feng, R.; Liu, J.; Huang, J.; Chen, Y.; Wang, X.; Sun, J.; et al. FGFR Blockade Boosts T Cell Infiltration into Triple-Negative Breast Cancer by Regulating Cancer-Associated Fibroblasts. Theranostics 2022, 12, 4564–4580. [CrossRef] [PubMed]

63. Kono, M.; Komatsuda, H.; Yamaki, H.; Kumai, T.; Hayashi, R.; Wakisaka, R.; Nagato, T.; Ohkuri, T.; Kosaka, A.; Ohara, K.; et al. Immunomodulation via FGFR Inhibition Augments FGFR1 Targeting T-Cell Based Antitumor Immunotherapy for Head and Neck Squamous Cell Carcinoma. Oncoimmunology 2022, 11, 2021619. [CrossRef] [PubMed]

64. Ocker, M. Fibroblast Growth Factor Signaling in Non-Alcoholic Fatty Liver Disease and Non-Alcoholic Steatohepatitis: Paving the Way to Hepatocellular Carcinoma. World J. Gastroenterol. 2020, 26, 279–290. [CrossRef] [PubMed]

65. Seitz, T.; Hellerbrand, C. Role of Fibroblast Growth Factor Signalling in Hepatic Fibrosis. Liver Int. 2021, 41, 1201–1215. [CrossRef]

66. Tsimafeyeu, I.; Ludes-Meyers, J.; Stepanova, E.; Daeyaert, F.; Khochenkov, D.; Joose, J.-B.; Solomko, E.; Van Akene, K.; Peretolchina, N.; Yin, W.; et al. Targeting FGFR2 with Alofanib (RPT835) Shows Potent Activity in Tumour Models. Br. J. Cancer 2021, 125, 1251–1260. [CrossRef]

67. Herbert, C.; Schieborr, U.; Saxena, K.; Juraszek, J.; De Smet, F.; Alcouffe, C.; Biancioletto, M.; Saladino, G.; Sibrac, D.; Kudlinzki, D.; et al. Molecular Mechanism of SSR128129E, an Extracellularly Acting, Small-Molecule, Allosteric Inhibitor of FGFR Receptor Signaling. Cancer Cell 2016, 30, 176–186. [CrossRef]

68. Loriot, Y.; Schuler, M.; Iyer, G.; Witt, O.; Doi, T.; Qin, S.; Taberner, J.; Reardon, D.A.; Massard, C.; Palmer, D.H.; et al. Tumor Agnostic Efficacy and Safety of Erdafitinib in Patients (Pts) with Advanced Solid Tumors with Prespecified Fibroblast Growth Factor Receptor Alterations (FGFRalt) in RAGNAR: Interim Analysis (IA) Results. J. Clin. Oncol. 2022, 40, 3007. [CrossRef]

69. Kurinovski, J.; Kameda, M.; Ikubo, K.; Hisamichi, H.; Kawamoto, Y.; Kikuchi, S.; Moritomo, H.; Terakawa, T.; Iwai, Y.; Noda, A.; et al. Discovery of ASP5878: Synthesis and Structure-Activity Relationships of Pyrimidine Derivatives as Pan-FGFRs Inhibitors with Improved Metabolic Stability and Suppressed HERG-Channel Inhibitory Activity. Bioorg. Med. Chem. 2022, 59, 116657. [CrossRef]

70. Yamamoto, N.; Ryoo, B.-Y.; Keam, B.; Kudo, M.; Lin, C.-C.; Kunieda, F.; Ball, H.A.; Moran, D.; Komatsu, K.; Takeda, K.; et al. A Phase 1 Study of Oral ASP5878, a Selective Small-Molecule Inhibitor of Fibroblast Growth Factor Receptors 1–4, as a Single Dose and Multiple Doses in Patients with Solid Malignancies. Invest. New Drugs 2020, 38, 445–456. [CrossRef]

71. Gavine, P.R.; Mooney, L.; Kilgour, E.; Thomas, A.P.; Al-Kadhimi, K.; Beck, S.; Rooney, C.; Coleman, T.; Baker, D.; Mellor, M.J.; et al. AZD4547: An Orally Bioavailable, Potent, and Selective Inhibitor of the Fibroblast Growth Factor Receptor Tyrosine Kinase Family. Cancer Res. 2012, 72, 2045–2056. [CrossRef]

72. Coombes, R.C.; Badman, P.D.; Lozano-Kuehne, J.P.; Liu, X.; Macpherson, I.R.; Zaubir, I.; Baird, R.D.; Rosenfeld, N.; Garcia-Corbacho, J.; Cresti, N.; et al. Results of the Phase Ia RADICAL Trial of the FGFR Inhibitor AZD4547 in Endocrine Resistant Breast Cancer. Nat. Commun. 2022, 13, 3246. [CrossRef] [PubMed]

73. Paik, P.K.; Shen, R.; Berger, M.F.; Ferry, D.; Soria, J.-C.; Mathewson, A.; Rooney, C.; Smith, N.R.; Cullberg, M.; Kilgour, E.; et al. A Phase Ib Open-Label Multicenter Study of AZD4547 in Patients with Advanced Squamous Cell Lung Cancers. Clin. Cancer Res. 2017, 23, 5366–5373. [CrossRef] [PubMed]
74. Chae, Y.K.; Hong, F.; Vaklavas, C.; Cheng, H.H.; Hammerman, P.; Mitchell, E.P.; Zwiebel, J.A.; Ivy, S.P.; Gray, R.J.; Li, S.; et al. Phase II Study of AZD4547 in Patients With Tumors Harboring Aberrations in the FGFR Pathway: Results From the NCI-MATCH Trial (EAY131) Subprotocol W. J. Clin. Oncol. 2020, 38, 2407–2417. [CrossRef]

75. Ebike, H.; Taka, N.; Matsumita, M.; Ohmori, M.; Takami, K.; Hyobohd, I.; Kohchi, M.; Hayase, T.; Nishii, H.; Morikami, K.; et al. Discovery of [5-Amino-1-(2-Methyl-3-Benzoimidazol-5-Yl)-Pyrazol-4-Yl]-1H-Indol-2-Yl]-Methane (CH5183284/Debio 1347), An Orally Available and Selective Fibroblast Growth Factor Receptor (FGFR) Inhibitor. J. Med. Chem. 2016, 59, 10586–10600. [CrossRef] [PubMed]

76. Voss, M.H.; Hierro, C.; Heist, R.S.; Cleary, J.M.; Meric-Bernstam, F.; Tabernero, J.; Janku, F.; Gandhi, L.; Iafrate, A.J.; Borger, D.R.; et al. A Phase I, Open-Label, Multicenter, Dose-Escalation Study of the Oral Selective FGFR Inhibitor Debio 1347 in Patients with Advanced Solid Tumors Harboring FGFR Gene Alterations. Clin. Cancer Res. 2019, 25, 2699–2707. [CrossRef] [PubMed]

77. Hall, T.G.; Yu, Y.; Eathiraj, S.; Wang, Y.; Savage, R.E.; Lapiere, J.-M.; Schwartz, B.; Abbadsada, G. Preclinical Activity of ARQ 087, a Novel Inhibitor of FGFR Dysregulation. PLoS ONE 2016, 11, e0162594. [CrossRef] [PubMed]

78. Mazzaferro, V.; El-Rayes, B.F.; Droz Dit Busset, M.; Cotsoglou, C.; Harris, W.P.; Damjanov, N.; Masi, G.; Rimassa, L.; Personeni, N.; Braiteh, F.; et al. Derazantinib (ARQ 087) in Advanced or Inoperable FGFR2 Gene Fusion-Positive Intrahepatic Cholangiocarcinoma. Br. J. Cancer 2019, 120, 165–171. [CrossRef] [PubMed]

79. Trudel, S.; Li, Z.H.; Wei, E.; Wiesmann, M.; Chang, H.; Chen, C.; Reece, D.; Heise, C.; Stewart, A.K. CHIR-258, a Novel, Multitargeted Tyrosine Kinase Inhibitor for the Potential Treatment of (4;14) Multiple Myeloma. Blood 2005, 105, 2941–2948. [CrossRef]

80. Escudier, B.; Grünwald, V.; Ravaud, A.; Ou, Y.-C.; Castellano, D.; Lin, C.-C.; Schewach, J.E.; Harzstark, A.; Beall, S.; Pirotta, N.; et al. Phase II Results of Dovitinib (TKI258) in Patients with Metastatic Renal Cell Cancer. Clin. Cancer Res. 2014, 20, 3012–3022. [CrossRef]

81. Watanabe Miyano, S.; Yamamoto, Y.; Kodama, K.; Miyajima, Y.; Mikamoto, M.; Nakagawa, T.; Kuramochi, H.; Funasaki, S.; Nagao, S.; Sugi, N.H.; et al. E7090, a Novel Selective Inhibitor of Fibroblast Growth Factor Receptors, Displays Potent Antitumor Activity and Prolongs Survival in Preclinical Models. Mol. Cancer Ther. 2016, 15, 2630–2639. [CrossRef]

82. Koyama, T.; Shimizu, T.; Iwasa, S.; Fujiwara, Y.; Kondo, S.; Kitano, S.; Yonemori, K.; Shimomura, A.; Izumi, S.; Sasaki, T.; et al. First-in-Human Phase I Study of E7090, a Novel Selective Fibroblast Growth Factor Receptor Inhibitor, in Patients with Advanced Solid Tumors. Cancer Sci. 2020, 111, 571–587. [CrossRef] [PubMed]

83. Chiba, Y.; Sudo, K.; Kojima, Y.; Okuma, H.; Kohsaka, S.; Machida, R.; Ichimura, M.; Anjo, K.; Kurishita, K.; Okita, N.; et al. A Multicenter Investigator-Initiated Phase 2 Trial of E7090 in Patients with Advanced or Recurrent Solid Tumor with Fibroblast Growth Factor Receptor (FGFR) Gene Alteration: FORTUNE Trial. BMC Cancer 2022, 22, 869. [CrossRef] [PubMed]

84. Perera, T.P.S.; Jovcheva, E.; Mevellec, L.; Vialard, J.; De Lange, D.; Verhulst, T.; Paulussen, C.; Van De Ven, K.; King, P.; Freyne, E.; et al. Discovery and Pharmacological Characterization of [N]-42756493 (Erdafitinib), a Functionally Selective Small-Molecule FGFR Family Inhibitor. Mol. Cancer Ther. 2017, 16, 1010–1020. [CrossRef]

85. Montazeri, K.; Bellmunt, J. Erdafitinib for the Treatment of Metastatic Bladder Cancer. Expert Rev. Clin. Pharmacol. 2020, 13, 1–6. [CrossRef]

86. Sootome, H.; Fujita, H.; Ito, K.; Ochiwa, H.; Fujioka, Y.; Ito, K.; Miura, A.; Sagara, T.; Ito, S.; Ohswa, H.; et al. Futibatinib Is a Novel Irreversible FGFR 1-4 Inhibitor That Shows Selective Antitumor Activity against FGFR-Deregulated Tumors. Cancer Res. 2020, 80, 4986–4997. [CrossRef] [PubMed]

87. Quinzii, A.; Zecchetto, C.; Casalino, S.; Gaule, M.; Pesoni, C.; Merz, V.; Contarelli, S.; Pietrobono, S.; Benhadji, K.A.; Melisi, D. Clinical Response to Futibatinib in Patients with High-Level FGFR2-Amplified Advanced Gastric Cancer: Two Case Reports. Clin. Drug Invest. 2022, 42, 697–701. [CrossRef] [PubMed]

88. Doi, T.; Shitara, K.; Kojima, T.; Kuboki, Y.; Matsubara, N.; Bando, H.; Yoh, K.; Naito, Y.; Hirai, K.; Hurokawa, Y.; et al. Phase I Study of the Irreversible FGFR1-Inhibitor Futibatinib in Japanese Patients with Advanced Solid Tumors. Cancer Sci. 2022. [CrossRef]

89. Mercier-Bernstam, F.; Bahlada, R.; Hierro, C.; Sanson, M.; Bridgewater, J.; Arkenau, H.-T.; Tran, B.; Kelley, R.K.; Park, J.O.; Javle, M.; et al. Futibatinib, an Irreversible FGFR1-4 Inhibitor, in Patients with Advanced Solid Tumors Harboring FGFR/FGFR2 Aberrations: A Phase I Dose-Expansion Study. Cancer Discov. 2022, 12, 402–415. [CrossRef]

90. Guagnano, V.; Furet, P.; Spanka, C.; Bordas, V.; Le Douget, M.; Stamm, C.; Brueggen, J.; Jensen, M.R.; Schnell, C.; Schmid, H.; et al. Discovery of 3-(2,6-Dichloro-3,5-Dimethoxy-Phenyl)-1-(6-[4-(4-Ethyl-Piperazin-1-Yl)-Pyrazin-4-Yl]-Pyrimidin-4-Yl)-1-Methyl-Urea (NVP-BGJ398), a Potent and Selective Inhibitor of the Fibroblast Growth Factor Receptor Family of Receptor Tyrosine Kinase. J. Med. Chem. 2011, 54, 7066–7083. [CrossRef]

91. Javle, M.; Lowery, M.; Shroff, R.T.; Weiss, K.H.; Springfeld, C.; Borad, M.J.; Ramanathan, R.K.; Goyal, L.; Sadeghi, S.; Macarulla, T.; et al. Phase II Study of BGJ398 in Patients With FGFR-Altered Advanced Cholangiocarcinoma. J. Clin. Oncol. 2018, 36, 276–282. [CrossRef] [PubMed]

92. Lassman, A.B.; Sepulveda-Sánchez, J.M.; Cloughesy, T.F.; Gil-Gil, M.J.; Puduvalli, V.K.; Raizer, J.J.; De Vos, F.Y.F.; Wen, P.Y.; Butowsk, N.A.; Clement, P.M.J.; et al. Infragitinib in Patients with Recurrent Gliomas and FGFR Alterations: A Multicenter Phase II Study. Clin. Cancer Res. 2022, 28, 2270–2277. [CrossRef] [PubMed]
93. Zhao, G.; Li, W.-Y.; Chen, D.; Henry, J.R.; Li, H.-Y.; Chen, Z.; Zia-Ebrahimi, M.; Bloem, L.; Zhai, Y.; Huss, K.; et al. A Novel, Selective Inhibitor of Fibroblast Growth Factor Receptors That Shows a Potent Broad Spectrum of Antitumor Activity in Several Tumor Xenograft Models. Mol. Cancer Ther. 2011, 10, 2200–2210. [CrossRef]

94. Michael, M.; Bang, Y.-J.; Park, Y.; Kang, Y.-K.; Kim, T.M.; Hamid, O.; Thornton, D.; Tate, S.C.; Raddad, E.; Tie, J. A Phase 1 Study of LY2874455, an Oral Selective Pan-FGFR Inhibitor, in Patients with Advanced Cancer. Target. Oncol. 2017, 12, 463–474. [CrossRef] [PubMed]

95. Holmström, T.H.; Moilanen, A.-M.; Ikonen, T.; Björkman, M.L.; Linnanen, T.; Wohlhaftrt, G.; Karlsson, S.; Oksala, R.; Korjamo, T.; Samajdar, S.; et al. ODM-203, a Selective Inhibitor of FGFR and VEGFR, Shows Strong Antitumor Activity, and Induces Antitumor Immunity. Mol. Cancer Ther. 2019, 18, 28–38. [CrossRef] [PubMed]

96. Bono, P.; Massard, C.; Pelitola, K.J.; Azaro, A.; Italiano, A.; Kristeleit, R.S.; Curigliano, G.; Lassen, U.; Arkenau, H.-T.; Hakulinen, P.; et al. Phase I/IIa, Open-Label, Multicentre Study to Evaluate the Optimal Dosing and Safety of ODM-203 in Patients with Advanced or Metastatic Solid Tumours. ESMO Open 2020, 5, e001081. [CrossRef]

97. Liu, P.C.C.; Koblish, H.; Wu, L.; Bowman, K.; Diamond, S.; DiMatteo, D.; Zhang, Y.; Hansbury, M.; Rupar, M.; Wen, X.; et al. INCBO354628 (Pemigatinib), a Potent and Selective Inhibitor of Fibroblast Growth Factor Receptors 1, 2, and 3, Displays Activity against Genetically Defined Tumor Models. PloS ONE 2020, 15, e0231877. [CrossRef]

98. Abou-Alfa, G.K.; Sahai, V.; Hollebecque, A.; Vaccaro, G.; Melisi, D.; Al-Rajabi, R.; Paulson, A.S.; Borad, M.J.; Gallinson, D.; Murphy, A.G.; et al. Pemigatinib for Previously Treated, Locally Advanced or Metastatic Cholangiocarcinoma: A Multicentre, Open-Label, Phase 2 Study. Cancer 2020, 116, 671–684. [CrossRef]

99. Gozgit, J.M.; Wong, M.J.; Moran, L.; Wardwell, S.; Mohemmad, Q.K.; Narasimhan, N.I.; Shakespeare, W.C.; Wang, F.; Clackson, T.; Rivera, V.M. Ponatinib (AP24534), a Multitargeted Pan-FGFR Inhibitor with Activity in Multiple FGFR-Amplified or Mutated Cancer Models. Mol. Cancer Ther. 2012, 11, 690–699. [CrossRef]

100. Ahn, D.H.; Uson Junior, P.L.S.; Masci, P.; Kosiorek, H.; Halfdanarson, T.R.; Mody, K.; Babiker, H.; DeLeon, T.; Sonbol, M.B.; Gores, G.; et al. A Pilot Study of Pan-FGFR Ponatinib in Patients with FGFR-Altered Advanced Cholangiocarcinoma. Investig. New Drugs 2022, 40, 134–141. [CrossRef]

101. Fairhurst, R.A.; Knoepfel, T.; Buschmann, N.; Leblanc, C.; Mah, R.; Todorov, M.; Nimsgern, P.; Ripoche, S.; Niklaus, M.; Warin, N.; et al. Discovery of Roblitinib (FGF401) as a Reversible-Covalent Inhibitor of the Kinase Activity of Fibroblast Growth Factor Receptor 4. J. Med. Chem. 2020, 63, 12542–12573. [CrossRef]

102. Collin, M.-P.; Lobell, M.; Hübsch, W.; Brohm, D.; Schirok, H.; Jautelat, R.; Lustig, K.; Bömer, U.; Vöhringer, V.; Helfrich, B.A.; et al. Discovery of Roblitinib (FGF401) as a Reversible-Covalent Inhibitor of the Kinase Activity of Fibroblast Growth Factor Receptor 4. J. Med. Chem. 2020, 63, 12542–12573. [CrossRef]

103. Smyth, E.C.; Turner, N.; Pearson, A.; Peckitt, C.; Chau, I.; Watkins, D.; Kilgour, E.; Smith, N.R.; Gillbanks, A.; Chua, S.; et al. FGFR1 Expression Levels Predict BGJ398 Sensitivity of FGFR1-Dependent Head and Neck Squamous Cell Cancers. Mol. Cancer Ther. 2011, 10, 671–684. [CrossRef] [PubMed]

104. Wynes, M.W.; Hinz, T.K.; Gao, D.; Martini, M.; Marek, L.A.; Ware, K.E.; Edwards, M.G.; Böhm, D.; Perner, S.; Helfrich, B.A.; et al. FGFR1 MRNA and Protein Expression, Not Gene Copy Number, Predict FGFR TKI Sensitivity across All Lung Cancer Histologies. Clin. Cancer Res. 2014, 20, 3299–3309. [CrossRef]

105. Smith, E.C.; Turner, N.; Pearson, A.; Peckitt, C.; Chau, I.; Watkins, D.; Kilgour, E.; Smith, N.R.; Gillbanks, A.; Chua, S.; et al. Phase II Study of AZD4547 in FGFR Amplified Tumors: Gastroesophageal Cancer (GC) Cohort Clinical and Translational Results. Clin. Cancer Res. 2015, 21, 4356–4364. [CrossRef] [PubMed]

106. Göke, F.; Fransen, A.; Hinz, T.K.; Marek, L.A.; Yoon, P.; Sharma, R.; Bode, M.; von Maassenhausen, A.; Lankat-Buttgereit, B.; Göke, A.; et al. FGFR1 Expression Levels Predict BGJ398 Sensitivity of FGFR1-Dependent Head and Neck Squamous Cell Cancers. Clin. Cancer Res. 2015, 21, 4356–4364. [CrossRef] [PubMed]

107. Bernard-Pierrot, I.; Brams, A.; Dunois-Lardé, C.; Caillault, A.; Diez de Medina, S.G.; Cappellen, D.; Graff, G.; Thiery, J.P.; Chopin, D.; Ricol, D.; et al. Oncogenic Properties of the Mutated Forms of Fibroblast Growth Factor Receptor 4. J. Med. Chem. 2020, 63, 437–445. [CrossRef]

108. Murphy, A.G.; et al. Pemigatinib for Previously Treated, Locally Advanced or Metastatic Cholangiocarcinoma. Mol. Cancer Ther. 2015, 14, 740–747. [CrossRef] [PubMed]

109. Wang, X.; Kim, Y.; Jeong, P.; Park, C.; Kim, W.T.; Ryu, D.H.; Cha, E.-J.; Ha, Y.-S.; Kim, T.-H.; Kwon, T.G.; et al. Expression Levels of FGFR3 as a Prognostic Marker for the Progression of Primary PT1 Bladder Cancer and Its Association with Mutation Status. Oncol. Lett. 2017, 14, 3817–3824. [CrossRef]

110. Guancial, E.A.; Werner, L.; Bellmunt, J.; Bamias, A.; Choueiri, T.K.; Ross, R.; Schutz, F.A.; Park, R.S.; O’Brien, R.J.; Hirsch, M.S.; et al. FGFR Expression in Primary and Metastatic Urothelial Carcinoma of the Bladder. Cancer Med. 2014, 3, 835–844. [CrossRef]

111. Jogo, T.; Nakamura, Y.; Shiota, K.; Bando, H.; Yasui, H.; Esaki, T.; Terazawa, T.; Sato, T.; Shinozaki, E.; Nishina, T.; et al. Circulating Tumor DNA Analysis Detects FGFR2 Amplification and Concurrent Genomic Alterations Associated with FGFR Inhibitor Efficacy in Advanced Gastric Cancer. Clin. Cancer Res. 2021, 27, 5619–5627. [CrossRef]

112. Wainberg, Z.A.; Enzinger, P.C.; Kang, Y.-K.; Yamaguchi, K.; Qin, S.; Lee, K.-W.; Oh, S.C.; Li, J.; Turk, H.M.; Teixeira, A.C.; et al. Randomized Double-Blind Placebo-Controlled Phase 2 Study of Bemarituzumab Combined with Modified FOLFOX6 (MFOLFOX6) in First-Line (1L) Treatment of Advanced Gastric/Gastroesophageal Junction Adenocarcinoma (FIGHT). J. Clin. Oncol. 2021, 39, 160. [CrossRef]

113. Schuler, M.; Nogova, L.; Heidenreich, A.; Tai, D.; Cassier, P.; Richly, H.; Cho, B.C.; Sayehli, C.M.; Bender, S.; Ocker, M.; et al. Anti-Tumor Activity of the Pan-FGFR Inhibitor Robagitnib in Patients with Advanced Urothelial Carcinomas Selected Based on Tumor FGFR MRNA Expression Levels. Ann. Oncol. 2017, 28, v295–v329. [CrossRef]
113. Kim, E.K.; Cho, Y.A.; Koh, Y.W.; Shin, H.A.; Cho, B.C.; Yoon, S.O. Prognostic Implications of Fibroblast Growth Factor Receptor 1 (FGFR1) Gene Amplification and Protein Overexpression in Hypopharyngeal and Laryngeal Squamous Cell Carcinoma. BMC Cancer 2020, 20, 348. [CrossRef] [PubMed]

114. Theelen, W.S.; Mittempergher, L.; Willems, S.M.; Bosma, A.J.; Peters, D.D.; van der Noort, V.; Japenga, E.J.; Peeters, T.; Koole, K.; Šustić, T.; et al. FGFR1, 2 and 3 Protein Overexpression and Molecular Aberrations of FGFR3 in Early Stage Non-Small Cell Lung Cancer. J. Pathol. Clin. Res. 2016, 2, 223–233. [CrossRef] [PubMed]

115. Tomiguchi, M.; Yamamoto, Y.; Yamamoto-Ibusuki, M.; Goto-Yamaguchi, L.; Fujiki, Y.; Fujiwara, S.; Sueta, A.; Hayashi, M.; Takeshita, T.; Inao, T.; et al. Fibroblast Growth Factor Receptor-1 Protein Expression Is Associated with Prognosis in Estrogen Receptor-Positive/Human Epidermal Growth Factor Receptor-2-Negative Primary Breast Cancer. Cancer Sci. 2016, 107, 491–498. [CrossRef]

116. Ahn, S.; Lee, J.; Hong, M.; Kim, S.T.; Park, S.H.; Choi, M.G.; Lee, J.-H.; Sohn, T.S.; Bae, J.M.; Kim, S.; et al. FGFR2 in Gastric Cancer: Protein Overexpression Predicts Gene Amplification and High-H Index Predicts Poor Survival. Mod. Pathol. 2016, 29, 1095–1103. [CrossRef]

117. Tokunaga, R.; Imamura, Y.; Nakamura, K.; Ishimoto, T.; Nakagawa, S.; Miyake, K.; Nakaji, Y.; Tsuda, Y.; Iwatsuki, M.; Baba, Y.; et al. Fibroblast Growth Factor Receptor 2 Expression, but Not Its Genetic Amplification, Is Associated with Tumor Growth and Worse Survival in Esophageal Gastric Junction Adenocarcinoma. Oncotarget 2016, 7, 19748–19761. [CrossRef]

118. Uson Junior, P.L.S.; DeLeon, T.T.; Bogenberger, J.M.; Pai, R.K.; Kostiorek, H.E.; Yin, J.; Ahn, D.H.; Sonbol, M.B.; Bekaii-Saab, T.; Mansfield, A.S.; et al. FGFR2-IIIb Expression by Immunohistochemistry Has High Specificity in Cholangiocarcinoma with FGFR2 Genomic Alterations. Dig. Dis. Sci. 2022, 67, 3979–3985. [CrossRef]

119. van Rhijn, B.W.G.; Mertens, L.S.; Mayr, R.; Bostrom, P.J.; Rea, F.X.; Zwarthoff, E.C.; Boormans, J.L.; Abas, C.; van Leenders, G.J.L.H.; Götz, S.; et al. FGFR2 Mutation Status and FGFR3 Expression in a Large Bladder Cancer Cohort Treated by Radical Cystectomy: Implications for Anti-FGFR3 Treatment? Eur. Urol. 2020, 78, 682–687. [CrossRef]

120. Moes-Sosnowska, J.; Skupinska, M.; Lechowicz, U.; Szczepulska-Wojcik, E.; Skronska, P.; Rozy, A.; Stepniewska, A.; Langfort, R.; Rudzinski, P.; Orlowski, T.; et al. FGFR1-4 RNA-Based Gene Alteration and Expression Assay in Squamous Non-Small Cell Lung Cancer. Int. J. Mol. Sci. 2022, 23, 10506. [CrossRef]

121. Dietrich, D. FGFR-targeted therapy in head and neck carcinomas. HNO 2021, 69, 172–184. [CrossRef]

122. Santolla, M.E.; Maggiolini, M. The FGF/FGFR System in Breast Cancer: Oncogenic Features and Therapeutic Perspectives. Cancers 2020, 12, 3029. [CrossRef] [PubMed]

123. De Luca, A.; Frezzetti, D.; Gallo, M.; Normanno, N. FGFR-Targeted Therapeutics for the Treatment of Breast Cancer. Expert Opin. Investig. Drugs 2017, 26, 303–311. [CrossRef] [PubMed]

124. Lee, S.J.; Hong, J.Y.; Kim, K.; Kim, K.-M.; Kang, S.Y.; Lee, T.; Kim, S.T.; Park, S.H.; Park, Y.S.; Lim, H.Y.; et al. Detection of Fusion Genes Using a Targeted RNA Sequencing Panel in Gastrointestinal and Rare Cancers. J. Oncol. 2020, 2020, 4659062. [CrossRef]

125. Gu, W.; Yang, J.; Wang, Y.; Xu, J.; Wang, X.; Du, F.; Hu, X.; Guo, H.; Song, C.; Tao, R.; et al. Comprehensive Identification of FGFR1-4 Alterations in 5557 Chinese Patients with Solid Tumors by next-Generation Sequencing. Am. J. Cancer Res. 2021, 11, 3893–3906. [PubMed]

126. Napolitano, A.; Ostler, A.E.; Jones, R.L.; Huang, P.H. Fibroblast Growth Factor Receptor (FGFR) Signaling in GIST and Soft Tissue Sarcomas. Cells 2021, 10, 1533. [CrossRef] [PubMed]

127. Chan, S.L.; Schuler, M.; Kang, Y.-K.; Yen, C.J.; Edeline, J.; Choo, S.P.; Lin, C.-C.; Okusaka, T.; Weiss, K.-H.; Macarulla, T.; et al. A First-in-Human Phase 1/2 Study of Combination and FGFR401 with Spartalizumab in Patients with Hepatocellular Carcinoma or Biomarker-Selected Solid Tumors. J. Exp. Clin. Cancer Res. CR 2022, 41, 189. [CrossRef] [PubMed]

128. Su, X.; Zhan, P.; Gavine, P.R.; Morgan, S.; Womack, C.; Ni, X.; Shen, D.; Bang, Y.-J.; Im, S.-A.; Ho Kim, W.; et al. FGFR2 Amplification Has Prognostic Significance in Gastric Cancer: Results from a Large International Multicentre Study. Br. J. Cancer 2020, 110, 967–975. [CrossRef]

129. Seo, A.N.; Jin, Y.; Lee, H.J.; Sun, P.-L.; Kim, H.; Jheeon, S.; Kim, K.; Lee, C.-T.; Chung, J.-H. FGFR1 Amplification Is Associated with Poor Prognosis and Smoking in Non-Small-Cell Lung Cancer. Virchows Arch. Int. J. Pathol. 2014, 465, 547–558. [CrossRef]

130. Hur, J.Y.; Chao, J.; Kim, Y.; Kim, K.-M.; Mlepner, S.J.; Lee, J. High-Level FGFR2 Amplification Is Associated with Poor Prognosis and Lower Response to Chemotherapy in Gastric Cancers. Pathol. Res. Pract. 2020, 216, 152878. [CrossRef]

131. Kuboki, Y.; Schatz, C.A.; Koechert, K.; Schubert, S.; Feng, J.; Wittmer-Rump, S.; Ziegler-Bäumer, K.; Krahn, T.; Nagaesuma, A.K.; Ochiai, A. In Situ Analysis of FGFR2 MRNA and Comparison with FGFR2 Gene Copy Number by Dual-Color in Situ Hybridization in a Large Cohort of Gastric Cancer Patients. Gastric Cancer 2018, 21, 401–412. [CrossRef]

132. Pearson, A.; Smyth, E.; Babina, I.S.; Herrera-Abreu, M.T.; Tarazona, N.; Peckitt, C.; Kilgour, E.; Smith, N.R.; Geh, C.; Rooney, C.; et al. High-Level Clonal FGFR Amplification and Response to FGFR Inhibition in a Translational Clinical Trial. Cancer Discov. 2016, 6, 838–851. [CrossRef] [PubMed]

133. Schildhaus, H.-U.; Heukamp, L.C.; Merkelbach-Bruse, S.; Riesner, K.; Schmitz, K.; Binot, E.; Pagen, E.; Albiss, K.; Schulte, W.; Ko, Y.-D.; et al. Definition of a Fluorescence In-Situ Hybridization Score Identifies High- and Low-Level FGFR1 Amplification Types in Squamous Cell Lung Cancer. Mod. Pathol. 2012, 25, 1473–1480. [CrossRef] [PubMed]

134. Kaibori, M.; Sakai, K.; Ishizaki, M.; Matsushima, H.; De Velasco, M.A.; Matsui, K.; Iida, H.; Kitade, H.; Kwon, A.-H.; Nagano, H.; et al. Increased FGF19 Copy Number Is Frequently Detected in Hepatocellular Carcinoma with a Complete Response after Sorafenib Treatment. Oncotarget 2016, 7, 49091–49098. [CrossRef]
Drilon, A.; Duruisseaux, M.; Han, J.-Y.; Ito, M.; Falcon, C.; Yang, S.-R.; Murciano-Goroff, Y.R.; Chen, H.; Okada, M.; Molina, Kazdal, D.; Hofman, V.; Christopoulos, P.; Ili_Goyal, L.; Shi, L.; Liu, L.Y.; Fece de la Cruz, F.; Lennerz, J.K.; Raghavan, S.; Leschiner, I.; Elagina, L.; Siravegna, G.; Ng, R.W.S.; Wolf, J.; Helland, Å.; Oh, I.-J.; Migliorino, M.R.; Dziadziuszko, R.; Wrona, A.; de Castro, J.; Mazieres, J.; Griesinger, F.; Chlistalla; Formisano, L.; Lu, Y.; Servetto, A.; Hanker, A.B.; Jansen, V.M.; Bauer, J.A.; Sudhan, D.R.; Guerrero-Zotano, A.L.; Croessmann, S.; Nogova, L.; Sequist, L.V.; Perez Garcia, J.M.; Andre, F.; Delord, J.-P.; Hidalgo, M.; Schellens, J.H.M.; Cassier, P.A.; Camidge, D.R.; S Geiss, G.K.; Bumgarner, R.E.; Birditt, B.; Dahl, T.; Dowidar, N.; Dunaway, D.L.; Fell, H.P.; Ferree, S.; George, R.D.; Grogan, T.; et al. High FGFR1-4 MRNA Expression Levels Correlate with Response to Selective FGFR Inhibitors in Breast Cancer. *Clin. Cancer Res.* **2022**, *28*, 137–149. [CrossRef]

Silverman, I.M.; Li, M.; Murugesan, K.; Krook, M.A.; Javele, M.M.; Kelley, R.K.; Borad, M.J.; Roychowdhury, S.; Meng, W.; Yilmaz, B.; et al. Validation and Characterization of FGFR2 Rearrangements in Cholangiocarcinoma with Comprehensive Genomic Profiling. *J. Mol. Diagn.* **2022**, *24*, 25–36. [CrossRef]

Freedman, A.N.; Klabunde, C.N.; Want, K.; Enewold, L.; Gray, S.W.; Filipski, K.K.; Keating, N.L.; Leonard, D.G.B.; Lively, T.; McNeel, T.S.; et al. Use of Next-Generation Sequencing Tests to Guide Cancer Treatment: Results From a Nationally Representative Survey of Oncologists in the United States. *JCO Precis. Oncol.* **2018**, *2*, 1–13. [CrossRef]

Vega, D.M.; Nishimura, K.K.; Zariffa, N.; Johnson, T.C.; Hoering, A.; Cilento, V.; Rosenthal, A.; Anagnostou, V.; Baden, J.; Beaver, J.A.; et al. Changes in Circulating Tumor DNA Reflect Clinical Benefit Across Multiple Studies of Patients With Non–Small-Cell Lung Cancer Treated With Immune Checkpoint Inhibitors. *JCO Precis. Oncol.* **2022**, *6*, e2100372. [CrossRef]

Duffy, M.J.; Crown, J. Circulating Tumor DNA as a Biomarker for Monitoring Patients with Solid Cancers: Comparison with Standard Protein Biomarkers. *Clin. Chem.* **2022**, hvac121. [CrossRef]

Mi, J.; Han, X.; Wang, R.; Ma, R.; Zhao, D. Circulation Tumour DNA in Predicting Recurrence and Prognosis in Operable Colorectal Cancer Patients: A Meta-Analysis. *Eur. J. Clin. Investig.* **2022**, e13842. [PubMed]

Goyal, L.; Saha, S.K.; Liu, L.Y.; Siravegna, G.; Leshchiner, I.; Ahronian, L.G.; Lennerz, J.K.; Vu, P.; Deshpande, V.; Kambadakone, A.; et al. Polyclonal Secondary FGFR2 Mutations Drive Acquired Resistance to FGFR Inhibition in Patients with FGFR2 Fusion-Positive Cholangiocarcinoma. *Cancer Discov.* **2017**, *7*, 252–263. [CrossRef] [PubMed]

Varghese, A.M.; Patel, J.; Janjigian, Y.Y.; Meng, F.; Selcuklu, S.D.; Iyer, G.; Houck-Loomis, B.; Harding, J.J.; O’Reilly, E.M.; Abou-Alfa, G.K.; et al. Noninnvasive Detection of Polyclonal Acquired Resistance to FGFR Inhibition in Patients With Cholangiocarcinoma Harboring FGFR2 Alterations. *JCO Precis. Oncol.* **2021**, *5*, 44–50. [CrossRef] [PubMed]

Goyal, L.; Shi, L.; Liu, L.Y.; Fece de la Cruz, F.; Lennerz, J.K.; Raghavan, S.; Leshchiner, I.; Elagina, L.; Siravegna, G.; Ng, R.W.S.; et al. TAS-120 Overcomes Resistance to ATP-Competitive FGFR Inhibitors in Patients with FGFR2 Fusion-Positive Intrahepatic Cholangiocarcinoma. *Cancer Discov.* **2019**, *9*, 1064–1079. [CrossRef]

Formisano, L.; Lu, Y.; Servetto, A.; Harker, A.B.; Jansen, V.M.; Bauer, J.A.; Sudhan, D.R.; Guerrero-Zotano, A.L.; Crossmann, S.; Guo, Y.; et al. aberrant FGFR signaling mediates resistance to cdk4/6 inhibitors in er+ breast cancer. *Nat. Commun.* **2019**, *10*, 1373. [CrossRef]

Nogova, L.; Sequist, L.V.; Perez Garcia, J.M.; Andre, F.; Delord, J.-P.; Hidalgo, M.; Schellens, J.H.M.; Cassier, P.A.; Camidge, D.R.; Schuler, M.; et al. Evaluation of BGJ398, a Fibroblast Growth Factor Receptor 1-3 Kinase Inhibitor, in Patients With Advanced Solid Tumors Harboring Genetic Alterations in Fibroblast Growth Factor Receptors: Results of a Global Phase I, Dose-Escalation and Dose-Expansion Study. *J. Clin. Oncol.* **2017**, *35*, 157–165. [CrossRef]

Miao, J.-L.; Zhou, J.-H.; Cai, J.-J.; Liu, R.-J. The Association between Fibroblast Growth Factor Receptor 1 Gene Amplification and Lung Cancer: A Meta-Analysis. *Eur. Arch. Med. Sci.* **2020**, *1*, 16–26. [CrossRef]

Quinn, D.L.; Petrylak, D.P.; Bellmunt, J.; Necchi, A.; Gurney, H.; Lee, J.-L.; Van der Heijden, M.S.; Rosenbaum, E.; Penel, N.; Pang, S.-T.; et al. FORT-I: Phase II/III Study of Rogaratinib versus Chemotherapy (CT) in Patients (Pts) with Locally Advanced or Metastatic Urothelial Carcinoma (UC) Selected Based on FGFR1/3 MRNA Expression. *J. Clin. Oncol.* **2020**, *38*, 489. [CrossRef]

Kazdal, D.; Hofman, V.; Christopoulos, P.; Ilie, M.; Stenzinger, A.; Hofman, P. Fusion-Positive Non-Small Cell Lung Carcinoma: Biological Principles, Clinical Practice, and Diagnostic Implications. *Genes. Chromosomes Cancer* **2022**, *61*, 244–260. [CrossRef]

Trombetta, D.; Sparaneo, A.; Fabrizio, F.P.; Di Micco, C.M.; Rossi, A.; Muscarella, L.A. NRG1 and NRG2 Fusions in Non-Small Cell Lung Cancer (NSCLC): Seven Years Between Lights and Shadows. *Expert Opin. Ther. Targets* **2021**, *25*, 865–875. [CrossRef]

Drilon, A.; Duruisseaux, M.; Han, J.-Y.; Ito, M.; Falcon, C.; Yang, S.-R.; Murciano-Goroff, Y.R.; Chen, H.; Okada, M.; Molina, M.A.; et al. Clinicopathologic Features and Response to Therapy of NRG1 Fusion-Driven Lung Cancers: The ENRGy1 Global Multicenter Registry. *J. Clin. Oncol.* **2019**, *37*, 2791–2802. [CrossRef] [PubMed]

Laetsch, T.W.; DuBois, S.G.; Mascarenhas, L.; Turpin, B.; Federman, N.; Albert, C.M.; Nagasubramanian, R.; Davis, J.L.; Rudzinski, E.; Feraco, A.M.; et al. Larotrectinib for Paediatric Solid Tumours Harbou ring NTRK Gene Fusions: Phase 1 Results from a Multicentre, Open-Label, Phase 1/2 Study. *Lancet Oncol.* **2018**, *19*, 705–714. [CrossRef]

Camidge, D.R.; Bang, Y.-J.; Kwak, E.L.; Iafrate, A.J.; Varela-Garcia, M.; Fox, S.B.; Riely, G.J.; Solomon, B.; Ou, S.-H.I.; Kim, D.-W.; et al. Activity and Safety of Crizotinib in Patients with ALK-Positive Non-Small-Cell Lung Cancer: Updated Results from a Phase 1 Study. *Lancet Oncol.* **2012**, *13*, 1011–1019. [CrossRef]

Wolf, J.; Helland, A.; Oh, I.-J.; Migliorino, M.R.; Dziedzicszko, R.; Wrona, A.; de Castro, J.; Mazieres, J.; Griesinger, F.; Chlitalla, M.; et al. Final Efficacy and Safety Data, and Exploratory Molecular Profiling from the Phase III ALUR Study of Alectinib versus Chemotherapy in Crizotinib-Pretreated ALK-Positive Non-Small-Cell Lung Cancer. *ESMO Open* **2022**, *7*, 100333. [CrossRef]

Prawira, A.; Le, T.B.U.; Ho, R.Z.W.; Huyhn, H. Uptregulation of the ErbB Family by EZH2 in Hepatocellular Carcinoma Confers Resistance to FGFR Inhibitor. *J. Cancer Res. Clin. Oncol.* **2021**, *147*, 2955–2968. [CrossRef]
157. Quintanal-Villalonga, A.; Molina-Pinelo, S.; Cirauqui, C.; Ojeda-Márquez, L.; Marrugal, Á.; Suarez, R.; Conde, E.; Ponce-Aix, S.; Enguita, A.B.; Carnero, A.; et al. FGFR1 Cooperates with EGFR in Lung Cancer Oncogenesis, and Their Combined Inhibition Shows Improved Efficacy. *J. Thorac. Oncol.* 2019, 14, 641–655. [CrossRef]

158. Datta, J.; Damodaran, S.; Parks, H.; Ocrainiciuc, C.; Miya, J.; Yu, L.; Gardner, E.P.; Samorodnitsky, E.; Wing, M.R.; Bhatt, D.; et al. Akt Activation Mediates Acquired Resistance to Fibroblast Growth Factor Receptor Inhibitor BGI398. *Mol. Cancer Ther.* 2017, 16, 614–624. [CrossRef]

159. Cowell, J.K.; Qin, H.; Hu, T.; Wu, Q.; Bhole, A.; Ren, M. Mutation in the FGFR1 Tyrosine Kinase Domain or Inactivation of PTEN Is Associated with Acquired Resistance to FGFR Inhibitors in FGFR1-Driven Leukemia/Lymphomas. *Int. J. Cancer* 2017, 141, 1822–1829. [CrossRef]

160. Lau, D.K.; Luk, I.Y.; Jenkins, L.J.; Martin, A.; Williams, D.S.; Schoffer, K.L.; Chionh, F.; Buchert, M.; Sjoquist, K.; Boussioutas, A.; et al. Rapid Resistance of FGFR-Driven Gastric Cancers to Regorafenib and Targeted FGFR Inhibitors Can Be Overcome by Parallel Inhibition of MEK. *Mol. Cancer Ther.* 2021, 20, 704–715. [CrossRef]

161. Siefker-Radtke, A.O.; Necchi, A.; Park, S.H.; García-Donas, J.; Huddart, R.A.; Burgess, E.F.; Fleming, M.T.; Rezazadeh Kalebasty, A.; Mellado, B.; Varlamov, S.; et al. Efficacy and Safety of Erdafitinib in Patients with Locally Advanced or Metastatic Urothelial Carcinoma: Long-Term Follow-up of a Phase 2 Study. *Lancet Oncol.* 2021, 23, 248–258. [CrossRef]

162. Vaclova, T.; Grazini, U.; Ward, L.; O’Neill, D.; Markovets, A.; Huang, X.; Chmielecki, J.; Hartmaier, R.; Thress, K.S.; Smith, P.D.; et al. Clinical Impact of Subclonal EGFR T790M Mutations in Advanced-Stage EGFR-Mutant Non-Small-Cell Lung Cancers. *Nat. Commun.* 2021, 12, 1780. [CrossRef] [PubMed]

163. Sevillano Fernández, E.; Madurga de Lacalle, R.; Rodríguez Moreno, J.F.; Barquín García, A.; Yagüe Fernández, M.; Navarro Alcaraz, P.; Barba Llacer, M.; Quiralte Pulido, M.; García-Donás Jiménez, J. Prognostic Value and Clinical Significance of FGFR Genomic Alterations (GAs) in Metastatic Urothelial Cancer Patients. *J. Clin. Med.* 2022, 11, 4483. [CrossRef] [PubMed]

164. Necchi, A.; Lo Vullo, S.; Raggi, D.; Gloghini, A.; Giannatempo, P.; Colecchia, M.; Mariani, L. Prognostic Effect of FGFR Mutations or Gene Fusions in Patients with Metastatic Urothelial Carcinoma Receiving First-Line Platinum-Based Chemotherapy: Results from a Large, Single-Institution Cohort. *Eur. Urol. Focus* 2019, 5, 853–856. [CrossRef] [PubMed]

165. Jing, W.; Wang, G.; Cui, Z.; Xiong, G.; Jiang, X.; Li, Y.; Li, W.; Han, B.; Chen, S.; Shi, B. FGFR3 Destabilizes PD-L1 via NEDD4 to Control T-Cell-Mediated Bladder Cancer Immune Surveillance. *Cancer Res.* 2022, 82, 114–129. [CrossRef] [PubMed]

166. Rosenberg, J.E.; Gajate, P.; Morales-Barrera, R.; Lee, J.-L.; Necchi, A.; Penel, N.; Zaganel, V.; Sierecki, M.R.; Bao, W.; Zhou, Y.; et al. Safety and Efficacy of Rogaratinib in Combination with Atezolizumab in Cisplatin-Ineligible Patients (Pts) with Locally Advanced or Metastatic Urothelial Cancer (UC) and FGFR MRNA Overexpression in the Phase Ib/II FORT-2 Study. *J. Clin. Oncol.* 2021, 39, 4521. [CrossRef]

167. Siefker-Radtke, A.O.; Loriot, Y.; Siena, S.; Beato, C.; Climent Duran, M.A.; Varlamov, S.; Duran, I.; Tagawa, S.T.; Geoffroi, L.; Mellado, B.; et al. Updated Data from the NORSE Trial of Erdafitinib (ERDA) plus Cetrelimab (CET) in Patients (Pts) with Metastatic or Locally Advanced Urothelial Carcinoma (MUC) and Specific Fibroblast Growth Factor Receptor (FGFR) Alterations. *Ann. Oncol.* 2020, 31, S584. [CrossRef]