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AVUTOMETINIB: A NOVEL RAF–MEK CLAMP INHIBITOR IN CANCER THERAPY – CHEMISTRY, MECHANISM, SAR, AND SIGNALING PATHWAY MODULATION

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ABSTRACT

The RAF–MEK–ERK signaling cascade is a central regulator of cell proliferation and survival, and its aberrant activation is a hallmark of many human cancers, particularly those harboring RAS or RAF mutations. While existing MEK and RAF inhibitors have demonstrated limited clinical success, their effectiveness is often compromised by feedback reactivation and acquired resistance. Avutometinib (VS-6766), a novel dual RAF–MEK clamp inhibitor, addresses these limitations by simultaneously inhibiting MEK activity and stabilizing it in an inactive complex with RAF, thereby preventing upstream feedback signaling and achieving sustained suppression of ERK activation. This unique mechanism disrupts the compensatory signaling often observed with classical MEK inhibitors. Preclinical studies demonstrate that avutometinib more effectively inhibits ERK phosphorylation, induces G1 arrest, and suppresses tumor growth in RAS-mutant cancer models compared to traditional pathway inhibitors. Early clinical trials, particularly in low-grade serous ovarian carcinoma (LGSOC) and KRAS-mutant non–small cell lung cancer (NSCLC), report encouraging response rates and durable disease control, especially when combined with focal adhesion kinase (FAK) inhibitors. These findings position avutometinib as a promising targeted therapy for RAS/MAPK-driven malignancies. Its mechanism offers a strategic advantage by overcoming resistance pathways that have historically limited MAPK-targeted therapies. Continued investigation into biomarker-driven patient selection and rational combination regimens is warranted to fully realize the clinical potential of avutometinib across diverse tumor types.

KEYWORDS: Cancer, chemistry, RAF-MEK-ERK, Signaling Pathway, Avutometinib

INTRODUCTION

The RAF–MEK–ERK Pathway in Cancer The RAF–MEK–ERK cascade (a core module of the MAPK pathway) is a critical signaling pathway that regulates cell

proliferation, survival, and differentiation. Aberrant activation of this pathway is a common driver of oncogenesis, occurring in a large fraction of human cancers. For example, roughly one-third of tumors have activating mutations in RAS genes, and about 8% harbor mutations in RAF kinases, leading to unchecked MAPK signaling and tumor growth. Given its central role in cancer cell biology, the RAS/RAF/MEK/ERK pathway has been a major focus of targeted therapy development over the past decade (1-3).

Kinase Inhibitors and Their Limitations

While RAF and MEK inhibitors have shown clinical success, their impact is often short-lived due to adaptive resistance. Tumor cells use feedback loops and signaling plasticity to bypass inhibition. A key issue is loss of negative feedback MEK inhibition can trigger rebound activation of RAF and RAS, reactivating ERK signaling. In RAS-mutant cancers, MEK inhibitors often lead to increased MEK phosphorylation, undermining their effectiveness (4, 5). Similarly, acquired resistance to RAF or MEK inhibitors frequently arises via secondary mutations or pathway re-routing, resulting in only transient responses in many patients. These challenges have spurred efforts to design combination therapies and next-generation inhibitors that can shut down MAPK signaling more durably by preventing compensatory pathway reactivation (1, 6).

Avutometinib as a Dual RAF–MEK Clamp Inhibitor

Avutometinib (VS-6766) is a novel kinase inhibitor designed to overcome resistance in MAPK pathway therapies. Unlike traditional MEK inhibitors, it functions as a "RAF–MEK clamp", simultaneously inhibiting MEK activity and locking it in an inactive complex with RAF. This dual action prevents RAF from reactivating MEK, effectively blocking feedback loops and achieving sustained ERK pathway suppression. Its unique mechanism makes Avutometinib the first-in-class molecule to target both RAF and MEK with a single agent (7, 8). Early studies showed that it can inhibit MAPK signaling more profoundly and for a longer duration than earlier MEK-only inhibitors (e.g. PD0325901) in preclinical models. Notably, this compound was initially identified through a screen for agents that induce the cell-cycle inhibitor p27, hinting at its distinctive signaling effects (9, 10).

Clinical Significance and Novelty

Avutometinib's dual inhibition strategy has significant clinical implications, positioning it as a novel therapeutic option for cancers driven by the RAS–RAF–MEK–ERK pathway. By blocking feedback loops and pathway escape routes, this agent has the potential to produce deeper and more durable responses in tumors that previously evaded single-node inhibitors. Clinically, avutometinib has shown promising activity in difficult-to-treat, MAPK-driven cancers. For example, in recurrent low-grade serous ovarian carcinoma – a cancer characterized by RAS/RAF mutations and relative resistance to standard chemotherapy – the combination of avutometinib with the focal adhesion kinase inhibitor defactinib achieved an objective response rate of approximately 42%, with a median progression-free survival around 20 months (5). This level of efficacy is notably higher than historical outcomes in that

disease, underscoring the potential of targeting the MAPK pathway in a more comprehensive manner. Avutometinib's early clinical success has spurred multiple ongoing trials in RAS- or BRAF-mutant solid tumors (and even RAS-driven multiple myeloma), including a registration-directed Phase II study in ovarian cancer (11). Taken together, these developments highlight why avutometinib is considered a novel agent: it introduces a first-of-its-kind mechanism that addresses the limitations of prior RAF or MEK inhibitors, and it has demonstrated encouraging efficacy in cancers with dysregulated MAPK signaling. As such, avutometinib represents an important advancement in the continued effort to therapeutically target the RAF–MEK–ERK pathway in cancer (12, 13).

Chemistry and SAR of Avutometinib

Avutometinib's journey from a screening hit to an optimized drug candidate is a tale of strategic medicinal chemistry and iterative design. It was originally discovered by researchers at Chugai Pharmaceutical (a Roche subsidiary) who started with a hit compound identified via high-throughput screening and then derivatized it to improve its properties. The result of this optimization campaign is Avutometinib (formerly CH5126766 or VS-6766), a molecule with a unique dual mechanism of action. Chemically, Avutometinib is a complex polycyclic compound – its IUPAC name (for the chemically inclined) is 3-[[3- fluoro-2-(methylsulfamoylamino)pyridin-4-yl]methyl]-4-methyl-7-pyrimidin-2-yloxychromen-2-one , and it has the molecular formula $C_{21}H_{18}FN_5O_5S$. In simpler terms, the molecule can be visualized as a coumarin core (a chromen-2-one scaffold) adorned with multiple heterocyclic rings and functional groups that were tactically added to enhance its activity and drug-like properties (14, 15).

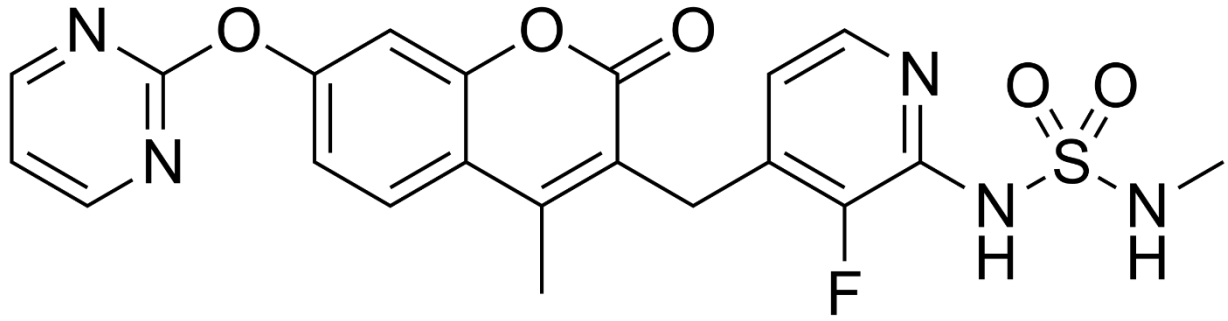

Figure 1: Chemical structure of Avutometinib (skeletal formula). Avutometinib features a chromen-2-one (coumarin) core with a 4-methyl substituent and a 7-pyrimidin-2-yloxy linkage, as well as a 3-[(pyridin-4-yl)methyl] substituent bearing a fluoro and methylsulfamoylamino group. These structural elements were fine-tuned by medicinal chemists to achieve potent inhibition of the Raf–MEK–ERK pathway.

The structure–activity relationship (SAR) development of Avutometinib is particularly fascinating. The initial lead compound, while showing promise as a MEK inhibitor, had suboptimal solubility and metabolic stability (16). Chemists embarked on a "nitrogen scan," systematically replacing certain carbon atoms in the molecular

scaffold with nitrogen heteroatoms to see how this affected potency and pharmacokinetics. This strategy stems from the idea that inserting nitrogen into aromatic rings can reduce lipophilicity and shield the molecule from metabolic enzymes, often improving solubility and stability. In the case of Avutometinib, these modifications were critical. For example, one key change was swapping a carbon for nitrogen in the pendant aromatic ring (converting a phenyl ring into a pyridine moiety). This single alteration – effectively changing a C–H to an N at a specific position – had a dramatic impact on the compound's drug-like profile (17).

To illustrate, an earlier analog (call it Compound 14) that lacked this nitrogen had excellent enzyme potency (slightly lower IC50 values) but was far less pharmaceutically optimized. Meanwhile, the version with the nitrogen (eventually Avutometinib, then known as Compound 1) achieved a far better balance between potency and pharmacokinetics. Table 1 compares these two variants, highlighting how strategic structural changes improved the molecule:

Table 1: Replacing the phenyl ring with a pyridine at X_3 slightly reduced in vitro potency but significantly improved solubility and pharmacokinetics. As a result, Compound 1 (Avutometinib) was chosen over Compound 14 for development (18).

Property	Compound 14 (early analog)	Compound 1 (Avutometinib)
Key structural difference	Phenyl ring (C–H at position X_3)	Pyridine ring (N at position X_3)
HCT116 cancer cell IC_{50}	17 nM (more potent)	40 nM (slightly less potent)
C-Raf enzyme IC_{50}	23 nM	56 nM
MEK1 enzyme IC_{50}	97 nM	160 nM
Aqueous solubility (pH 7)	13 µg/mL	159 µg/mL
Metabolic stability (liver microsomes)	CLint = 6.7 µL/min/mg (rapid clearance)	CLint = 0.5 µL/min/mg (much slower clearance)
Oral exposure (mouse AUC)	425 µM·h	2831 µM·h

Another notable structural feature is the methylsulfamoylamino group on the pyridine ring (–NH– SO_2–CH_3). This sulfonamide group not only adds polarity (aiding solubility) but also can engage in hydrogen bonding. In fact, modeling suggested that an intramolecular hydrogen bond involving this sulfamide helps lock the molecule in a bioactive conformation . The presence of a fluorine atom on that same ring (at the 3-position of the pyridine) is a classic medicinal chemistry tactic to block an undesirable metabolism hotspot and stabilize the molecule further. All these tweaks – the fluorine, the sulfonamide, the heterocyclic inserts – collectively fine-tuned Avutometinib's pharmacological profile (19, 20).

Avutometinib's unique bifunctional design enables it to act as a "RAF/MEK clamp," preventing RAF reactivation and overcoming feedback seen with traditional MEK inhibitors. Its coumarin–pyrimidine scaffold binds an allosteric MEK pocket, inducing an inactive MEK–RAF complex. This dual action blocks MEK activity and locks RAF, effectively shutting down the Ras–Raf–MEK–ERK pathway in RAS-driven cancers (21).

Mechanism of Action

RAF–MEK–ERK Signaling Cascade

The anti-tumor effects of avutometinib are best understood in the context of the RAF–MEK–ERK signaling cascade. In normal mitogenic signaling, activated RAS GTPases recruit RAF kinases (A-RAF, B-RAF, C-RAF) to the cell membrane, where RAF phosphorylates and activates MEK (MAPK/ERK kinase) on two key serine residues in MEK's activation loop (22). Once phosphorylated, MEK in turn phosphorylates ERK (extracellular signal–regulated kinase), propagating the signal to nuclear targets and driving cell proliferation and survival. This RAF–MEK–ERK module is tightly regulated by feedback mechanisms: active ERK phosphorylates upstream components (including RAF and receptor adaptors) to dampen excessive signaling and maintain homeostasis. In cancers with RAS or RAF mutations, however, this pathway becomes constitutively active and a critical driver of tumor growth, making its components prime therapeutic targets (16, 23).

Feedback Activation with Classical MEK Inhibitors

Conventional allosteric MEK inhibitors, such as trametinib and selumetinib, bind to MEK and prevent its kinase activity, but they do not prevent RAF from associating with and phosphorylating MEK. In fact, by blocking ERK output, these drugs relieve ERK-dependent negative feedback on RAF, often leading to a compensatory hyper-activation of RAF (24, 25). As a result, RAF phosphorylates MEK at elevated levels – a paradoxical increase in MEK phosphorylation (pMEK) despite the presence of a MEK inhibitor. This feedbackdriven pMEK induction has been observed with first-generation MEK inhibitors (e.g. PD0325901 or selumetinib) in RAS-mutant tumor models (26). Although MEK's kinase activity remains inhibited by the drug, the accumulation of phosphorylated MEK is a hallmark of the pathway's attempt to reactivate. Importantly, this compensatory RAF activation can reactivate upstream signaling networks and lessen the depth and durability of ERK pathway suppression achieved by classical MEK inhibitors. In clinical contexts, such feedback limits the efficacy of MEK inhibitor monotherapy, as tumors can escape via rebound MAPK pathway activity (27, 28).

Avutometinib's Novel "RAF–MEK Clamp" Mechanism

Avutometinib (VS-6766) was rationally designed to overcome these feedback limitations through a unique "RAF–MEK clamp" mechanism. Unlike traditional MEK inhibitors, avutometinib allosterically binds MEK in a way that stabilizes the MEK–RAF interaction and locks both proteins in an inactive complex . Structural and biochemical studies indicate that when avutometinib occupies MEK, it induces a MEK conformation that cannot be efficiently phosphorylated by RAF, nor released

from the RAF–MEK complex . In essence, the drug-bound MEK acts as a dominant-negative scaffold that sequesters RAF in a dormant state (29). This "clamping" of RAF and MEK serves two coordinated functions: (1) MEK inhibition – avutometinib directly blocks MEK's kinase activity toward ERK, and (2) RAF inhibition by complex – the trapped RAF cannot phosphorylate other MEK molecules or propagate signals. Avutometinib thus prevents MEK from being activated by RAF, in contrast to classical MEK inhibitors which permit RAF to continually phosphorylate MEK. Notably, because RAF is bound in an inactive MEK complex, avutometinib functionally suppresses RAF kinase activity upstream as well (30) . This dual action underlies its designation as a first-in-class "RAF/MEK clamp" inhibitor. Cellular studies have confirmed that avutometinib treatment drives the formation of stable, inert RAF–MEK complexes (sometimes termed dominant-negative complexes) that block RAF-dependent MEK phosphorylation across all RAF isoforms (A-RAF, B-RAF, and CRAF) (28, 31).

Suppression of ERK Phosphorylation and Feedback Loops

Avutometinib delivers a completer and more durable shutdown of the MAPK pathway by clamping RAF–MEK and blocking MEK activation. This prevents ERK phosphorylation downstream and disrupts compensatory feedback loops upstream. Unlike traditional MEK inhibitors, avutometinib does not trigger a rise in pMEK levels, meaning RAF remains under ERK-dependent feedback control. As a result, the pathway is insulated from reflex reactivation—a common limitation of single-node inhibitors. This dual mechanism ensures deeper, sustained ERK suppression and enhances antiproliferative efficacy in RAS/RAF-driven cancers (28).

Evidence from Preclinical Studies

Extensive preclinical evidence supports this dual MEK/RAF inhibition mechanism of avutometinib. In RASmutant cell lines, standard MEK inhibitors (e.g. selumetinib) cause a rebound increase in pMEK, whereas avutometinib completely abrogates this effect . For instance, Wada et al. (2014) (32) demonstrated that CH5126766 (avutometinib) blocked the MEK re-phosphorylation that occurred after selumetinib exposure in KRAS- or NRAS-mutant cancer cells. Correspondingly, avutometinib-treated cells showed more complete ERK pathway inhibition and greater anti-proliferative effects than cells treated with a classical MEK inhibitor . In the same study, avutometinib induced G1 cell-cycle arrest and apoptosis markers more effectively, consistent with more robust ERK suppression (32).

In in vivo models, the RAF–MEK clamp mechanism has translated into potent antitumor activity. Ishii et al. (2013) (28) reported that CH5126766 did not drive MEK hyperphosphorylation in tumor xenografts, yet achieved stronger ERK pathway blockade and tumor growth inhibition compared to a traditional MEK inhibitor . Similarly, in a NRAS mutant melanoma xenograft (SK-MEL-2), avutometinib caused significant tumor regression without the dose-limiting toxicities seen at equivalent doses of earlier MEK inhibitors . These data indicate that by preventing feedback reactivation of RAF, avutometinib maintains a suppressive grip on the MAPK cascade, leading to superior downstream signaling suppression and tumor control.

Notably, this clamp mechanism appears especially beneficial in RAS-mutant contexts, where upstream feedback activation is a prominent resistance mechanism – indeed, avutometinib has shown efficacy in KRAS-driven lung and ovarian cancer models where single-pathway inhibitors alone were less effective (28).

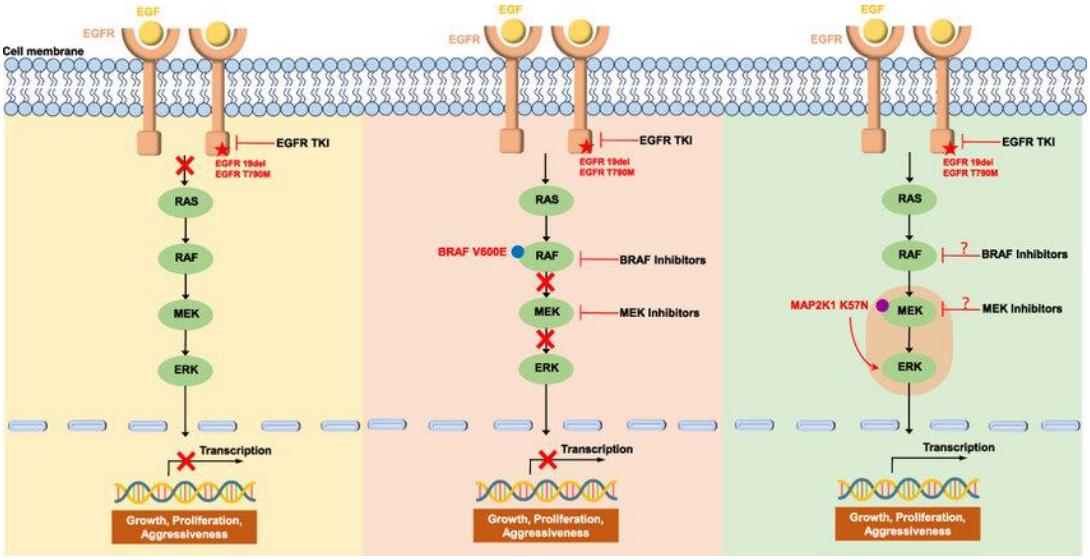

Figure 2. A schematic diagram of the RAF–MEK–ERK pathway is included to illustrate avutometinib's mechanism of action in contrast to classical MEK inhibitors.

Table 2: Comparative mechanistic and preclinical profiles of Avutometinib versus classical MEK inhibitors (31-33).

Aspect	**Avutometinib (RAF–MEK Clamp)**	**Classical MEK Inhibitors *(e.g., Selumetinib, Trametinib)***
Mechanism	Dual RAF–MEK inhibitor; locks RAF–MEK in inactive state	MEK-only inhibitor; does not block RAF
pMEK Levels	No increase; MEK cannot be phosphorylated	High increase due to feedback from ERK inhibition
pERK Suppression	Strong and sustained	Initial drop, but rebound common in RAS-mutant tumors
Feedback Loop Control	Effectively blocked	Feedback reactivation occurs
RAF–MEK Complex	Forms dominant-negative complex; RAF held inactive	No complex formed; RAF can reactivate MEK
Tumor Response (RAS-mutant models)	Tumor regression observed	Mostly halts growth; rarely causes regression
Cell Effect	G1 arrest; low apoptosis as single agent	G1 arrest; low apoptosis

Tolerability (Preclinical)	Well-tolerated; minimal side effects	Dose-limiting toxicities (eye, skin, GI) common

Signaling Pathway Modulation

RAF–MEK–ERK Pathway Inhibition and Feedback Suppression

Avutometinib is a first-in-class RAF–MEK "clamp" that uniquely modulates the MAPK pathway by simultaneously targeting MEK1/2 and upstream RAF kinases . Unlike conventional MEK inhibitors, which often relieve ERK-mediated feedback on RAF and trigger a compensatory rise in MEK phosphorylation, avutometinib stabilizes MEK in an inactive complex with RAF (34, 35). By binding to MEK and inducing a conformational change, avutometinib prevents RAF (including A-RAF, B-RAF, and C-RAF) from rephosphorylating and reactivating MEK . This dual mechanism leads to sustained ERK pathway inhibition without the rebound increase in MEK phosphorylation observed with other inhibitors (36, 37). Indeed, CH5126766 (the research precursor to avutometinib) was shown to avoid RAF-driven pMEK induction and achieved deeper suppression of ERK signaling output than a standard allosteric MEK inhibitor in RAS-mutant tumor models. By effectively clamping the RAF–MEK module in an inactive state, avutometinib blocks the propagation of mitogenic signals through ERK and mitigates the immediate feedback loops that often undermine MAPK pathway inhibitors (28) .

Table 3 – Comparison of Avutometinib and Other MAPK Pathway Inhibitors: Signaling Impact & Resistance Mechanisms (5, 28, 31)

Parameter	**Avutometinib (RAF–MEK Clamp)**	**Allosteric MEK Inhibitor *(e.g., Trametinib)***	**BRAFV600E Inhibitor *(e.g., Vemurafenib)***
Mechanism	Dual RAF–MEK blocker; locks both in inactive complex	MEK-only blocker; no RAF inhibition	Blocks mutant BRAFV600E; no effect on wild-type RAF
pMEK Feedback	No increase; feedback loop blocked	Strong pMEK rise due to ERK feedback loss	Decreased in BRAF-mutants; increased in RAS-mutants (paradoxical activation)
ERK Suppression	Strong & sustained; no rebound	Initial drop, often rebounds	Effective in BRAF-mutants; may activate ERK in RAS-mutants
RAS-Mutant Tumors	Effective; induces tumor regression	Limited activity; often needs combos	Ineffective or harmful due to feedback activation
Resistance	Minimal feedback; resistance via alternate pathways (e.g., PI3K/FAK)	Reactivation of pathway or escape mutations	Many routes: NRAS/MEK mutations, RTK/PI3K activation; combos delay resistance

Impact in KRAS-Mutant and RAS-Altered Tumors

Because of its mechanism, avutometinib is particularly effective in cancers driven by mutations in KRAS, NRAS, or other RAS–pathway alterations. RAS-mutant tumor cells are often highly dependent on the RAF– MEK–ERK cascade for proliferation, yet they rapidly adapt to single-agent MEK or RAF inhibitors via pathway reactivation. Avutometinib's ability to clamp the pathway shut has translated into promising antitumor activity in preclinical models and early clinical trials: - Preclinical Findings: In vitro, avutometinib suppresses ERK signaling and cell growth across multiple KRAS/NRAS-mutant cancer cell lines (including pancreatic, colon, lung, and melanoma models) (38). It induces G1-cell cycle arrest and pro-apoptotic changes in RAS-driven melanoma cells, accompanied by upregulation of CDK inhibitor p27 and downregulation of cyclin D1 (39, 40). Notably, CH5126766 (VS-6766) outperformed standard MEK inhibitors in RAS-mutant settings – for example, it more effectively reduced colony formation in NRAS- and KRAS-mutant cells compared to a MEK-only inhibitor, consistent with its enhanced ERK blockade (32). In vivo, single-agent avutometinib has shown tumor regression or growth delay in RAS-mutant xenograft models (e.g. KRAS^G12D colorectal and NRAS-mutant melanoma), whereas comparators had minimalimpact . These data underscore that RAS-mutant tumors, traditionally resistant to MAPK inhibition, become susceptible when both MEK and RAF feedback are co-targeted.

Low-grade serous ovarian cancer (LGSOC) is a slow-growing but treatment-resistant tumor, with ~30% harboring KRAS mutations and others showing MAPK pathway alterations. Traditional MEK inhibitors show limited clinical benefit (15–26% response rates). In contrast, early data on Avutometinib demonstrates significantly improved outcomes in RAS-altered LGSOC (41). In the Phase 1 FRAME trial (an exploratory study of avutometinib plus the FAK inhibitor defactinib), patients with recurrent LGSOC achieved an ORR of ~42% with the combination . Median progression-free survival (mPFS) reached 20.1 months , which is strikingly prolonged relative to historical controls (mPFS ~9–13 months on prior MEK inhibitors) . Importantly, responses were enriched in KRASmutant LGSOC: among patients with confirmed KRAS mutations, ORR was ~58% and mPFS ~30.8 months (31). These results, published in 2025, represent one of the highest response rates seen in this disease, and directly led to breakthrough therapy designation and the design of registrational trials . The subsequent randomized Phase 2 RAMP 201 trial confirmed an ORR of ~44% for avutometinib + defactinib in KRAS-mutant LGSOC, supporting an FDA accelerated approval in this molecular subset . Notably, some activity was observed even in KRAS wild-type LGSOC, suggesting the combo can overcome non-KRAS MAPK pathway drivers as well (42). Overall, in ovarian cancer models, avutometinib's dual pathway blockade – especially when combined with FAK inhibition – has proven capable of inducing tumor shrinkage and durable disease control in a setting where conventional therapies rarely succeed (43).

KRAS-Mutant NSCLC and Avutometinib: KRAS mutations are present in ~25–30% of non–small cell lung cancers (NSCLC), and MEK inhibitors have shown limited success in this setting. However, early studies show that Avutometinib

achieves ERK pathway suppression and modest activity in KRAS-mutant NSCLC. In the FRAME trial, single-agent avutometinib induced partial responses in some heavily pretreated patients. When combined with **defactinib**, the overall response rate (ORR) reached ~11%, including responses in $KRAS^{G12V}$ and $KRAS^{G12C}$ (44, 45). Though the response rate was modest, these results provided a proof-of-concept that dual RAF/MEK and FAK targeting can overcome some resistance in KRAS-driven lung tumors. Notably, disease control was seen in $KRAS^{G12V}$ NSCLC, a genotype not targetable by current $KRAS^{G12C}$ inhibitors. This has spurred further trials: for instance, the ongoing RAMP 202 study is evaluating avutometinib ± defactinib specifically in $KRAS^{G12V}$ mutant NSCLC, an area of unmet need (46).

Colorectal and Pancreatic Cancers: RAS-pathway alterations are prevalent in colorectal cancer (CRC) and pancreatic ductal adenocarcinoma (PDAC), but these tumors have proven extremely difficult to treat with MAPK inhibitors due to redundant survival signals. In the FRAME trial, RASmutant CRC and PDAC cohorts did not achieve confirmed responses with avutometinib + defactinib dual therapy , indicating that additional mechanisms drive resistance in these diseases. However, the trial's pharmacodynamic data confirmed effective ERK pathway inhibition in tumor biopsies (47, 48) . Building on these findings, new combination trials have been launched to tackle compensatory pathways: for example, avutometinib + defactinib is being combined with standard chemotherapy in PDAC (VS-6766 + gemcitabine/nab-paclitaxel) and with EGFR inhibition in CRC (VS-6766 + cetuximab) . The rationale is that vertical blockade of MAPK signaling (RAF/MEK ± EGFR) plus concomitant targeting of parallel survival signals can produce synergistic anti-tumor effects. Preclinically, there is evidence that dual MAPK and EGFR/RTK inhibition is beneficial in RAS-driven CRC , and that adding MEK or ERK inhibitors can overcome feedback resistance to $KRAS^{G12C}$ inhibitors in CRC models (49). Thus, while single-agent avutometinib has limited efficacy in tumors with highly redundant signaling (like KRAS-mutant CRC/PDAC), it serves as a potent backbone for combination regimens aiming to shutdown the RAS–ERK axis alongside other nodes.

Preclinical and Clinical Studies of Avutometinib

Preclinical Efficacy in RAS Pathway–Altered Cancers

Extensive preclinical studies have demonstrated that avutometinib (VS-6766) is active against a variety of RAS/MAPK pathway–driven tumors. As a dual RAF/MEK inhibitor or "RAF/MEK clamp," avutometinib potently suppresses MEK→ERK signaling while preventing upstream feedback reactivation of RAF (50, 51). In vitro, single-agent avutometinib inhibits proliferation across multiple cancer cell lines bearing KRAS, BRAF, or NF1 mutations. For example, in KRAS-mutant non–small cell lung cancer (NSCLC) models, avutometinib monotherapy showed cytotoxic activity in culture and tumor growth inhibition in xenografts . However, parallel activation of compensatory pathways often limits the depth and durability of MEK pathway inhibition by single agents (52, 53). In vivo, intermittent dosing schedules of avutometinib have been optimized to enhance efficacy while mitigating toxicity: preclinical pharmacology supported twiceweekly dosing, which was later validated clinically as sufficient to maintain ERK pathway suppression with improved

tolerability . This intermittent schedule distinguishes avutometinib from traditional daily MEK inhibitors and was critical for chronic administration in subsequent trials (54) .

Synergy of Avutometinib with Combination Therapies Combination strategies have been pursued to overcome adaptive resistance mechanisms that emerge with MAPK pathway blockade. Notably, focal adhesion kinase (FAK) activation has been identified as a key bypass mechanism when MEK is inhibited (55). In KRAS-driven tumors, MEK inhibition by avutometinib can lead to upregulation of FAK–RhoA signaling, promoting cell survival and metastasis . Addition of FAK inhibitors (e.g. defactinib or VS-4718) synergistically enhances avutometinib's anti-tumor effects by blocking this resistance pathway (56, 57). In vitro, the combination of avutometinib plus defactinib produces greater apoptosis and growth inhibition than either agent alone in RAS/MAPK-mutant cancer cell lines (53). For instance, five of five primary high-grade endometrioid endometrial cancer cell lines (several harboring KRAS, BRAF, or PIK3CA mutations) were sensitive to single-agent avutometinib, and all showed further reduced viability with the addition of a FAK inhibitor . Western blot analyses confirmed that the drug combination more completely suppresses downstream signaling (reducing phosphorylated ERK and MEK) while also decreasing p-FAK levels, consistent with dual-pathway blockade (58).

Preclinical in vivo findings mirror these synergistic effects. In KRAS-driven tumor xenografts, avutometinib plus defactinib yielded superior tumor regression compared to either monotherapy. For example, in mice engrafted with an aggressive RAS-mutated endometrial carcinoma (UTE10), the combination achieved significantly greater tumor growth inhibition than single agents (with tumor shrinkage evident by Day 9, $p<0.05$) (53). In BRAF V600E mutant melanoma models resistant to standard BRAF+MEK inhibitor therapy, the addition of a FAK inhibitor similarly restored sensitivity: avutometinib combined with a FAK inhibitor overcame acquired resistance to both targeted BRAF/MEK therapy and immune checkpoint blockade, yielding durable tumor regressions in vivo (59). Collectively these studies provide a strong mechanistic rationale for combining avutometinib with FAK inhibitors to achieve more complete and durable RAS/MAPK pathway suppression by blocking known escape pathways (49). This preclinical evidence has directly informed clinical trial design, especially for RAS-driven solid tumors where adaptive resistance limits single-agent efficacy.

Early-Phase Clinical Trials of Avutometinib Monotherapy

Initial first-in-human studies of avutometinib explored its tolerability and single-agent activity in patients with refractory solid tumors harboring RAS/RAF pathway alterations . In a phase I dose-escalation trial (including a "basket" expansion in RAS-mutant tumors), an intermittent dosing schedule was implemented based on preclinical insights (31). Avutometinib given orally twice weekly (3 weeks on, 1 week off) was generally well tolerated up to 4 mg per dose; however, a higher continuous dose or daily schedule proved too toxic, manifesting as dose-limiting rash and creatine kinase (CK) elevation. The recommended phase 2 dosing (RP2D) was established as 3.2 mg

twice weekly on the 3-weeks-on/1-week-off schedule . Even as monotherapy, preliminary anti-tumor activity was observed: partial responses occurred in heavily pretreated patients with KRAS^G12D-mutant low-grade serous ovarian cancer (LGSOC) and NRAS-mutant thyroid cancer, among others (60). In KRAS-mutant NSCLC, single-agent avutometinib achieved stable disease or minor tumor regressions in a subset of patients . These early signals of efficacy (albeit modest) validated that intermittent avutometinib could engage the target in RAS-driven cancers and paved the way for combination trials aiming to improve response rates (61).

Avutometinib + Defactinib Combination: FRAME Trial (Phase I)

The Phase I FRAME trial tested Avutometinib + Defactinib in RAS/RAF-mutant solid tumors, including NSCLC, CRC, pancreatic cancer, and LGSOC. An intermittent dosing schedule (Avutometinib 3.2 mg BID + Defactinib 200 mg BID, 3 weeks on/1 week off) was established as the RP2D due to better tolerability. Higher doses increased rash and fatigue. At RP2D, the combo showed a manageable safety profile, with common side effects including mild rash (~90%), elevated CK (56%), and rare grade 3–4 events. No unexpected safety concerns were observed (31).

Despite some required dose interruptions for chronic rash, the combination demonstrated encouraging efficacy in several RAS-driven cancers. The most striking activity was observed in low-grade serous ovarian cancer. Among 26 evaluable patients with KRAS-mutant LGSOC treated on FRAME, the confirmed objective response rate (ORR) was 42.3% (11 of 26, 95% CI ~23–63%) (62). In LGSOC, the combination achieved deep and lasting responses, with a median PFS of 20.1 months, significantly outperforming historical MEK inhibitor monotherapy (e.g., trametinib PFS ~13 months, ORR ~26%). Responses were seen across multiple RAS mutations ($KRAS^{G12D}$ and $KRAS^{G12V}$) and even in BRAF-mutant gynecologic tumors, highlighting the regimen's broad activity (31).

Efficacy in other FRAME cohorts was less pronounced, underscoring tumor-specific differences. In KRASmutant NSCLC, the combination achieved an ORR of ~11.1% with a median PFS of only ~3.5 months . A few NSCLC patients did respond (partial responses were confirmed in tumors with KRAS^G12V and KRAS^G12C mutations), but the overall response rate was relatively low . Likewise, no confirmed responses were seen in the small cohorts of pancreatic adenocarcinoma or KRAS/NRAS-mutant colorectal cancer treated with avutometinib ± defactinib (63). Many of those heavily pretreated pancreatic and CRC patients did achieve stable disease, and the combination proved tolerable in these groups . However, the lack of objective responses suggested that dual RAF/MEK+FAK inhibition may be insufficient alone in certain aggressive RAS-driven malignancies (especially those like pancreatic cancer, where parallel PI3K or other pathways contribute to growth). As a result, follow-up studies were launched to combine avutometinib with standard chemotherapies in these diseases – for example, adding it to gemcitabine/nab-paclitaxel in pancreatic cancer – to determine if a triplet approach can improve efficacy (49).

Phase II Clinical Outcomes in Key Indications

Low-Grade Serous Ovarian Cancer (RAMP 201)

Given the promising signals in LGSOC, a registration-directed phase II trial (RAMP 201) was undertaken to further evaluate avutometinib ± defactinib in this setting . Low-grade serous ovarian carcinoma is a slow-growing but therapy-resistant malignancy frequently driven by KRAS or BRAF mutations. RAMP 201 enrolled patients with recurrent LGSOC after ≥1 prior systemic therapy, stratifying by KRAS mutation status (64). In the initial randomized phase, patients received either avutometinib alone or in combination with defactinib (at two dose levels). The combination showed significantly better outcomes, with a 45% ORR vs. only 0–4% with monotherapy.

- KRAS-mutant LGSOC: ORR 60%
- KRAS wild-type: ORR ~29%

Notably, responses occurred in patients previously resistant to MEK inhibitors, suggesting the combo may overcome prior resistance. These strong results supported trial expansion and regulatory approval efforts (65).

In the full expansion of RAMP 201 (n=109), the confirmed ORR was 31%, including 2% complete and 29% partial responses.

- KRAS-mutant tumors:
  - ORR 44%
  - Median PFS: 22.0 months
  - Median Duration of Response (DOR): 31.1 months
- KRAS wild-type tumors:
  - ORR 17%
  - Median PFS: 12.8 months
  - Median DOR: 9.2 months

These results highlight the critical role of biomarker selection, with the combination showing greatest efficacy in RAS pathway-altered LGSOC. However, KRAS wild-type patients still benefited, possibly due to other MAPK-related alterations.

In RAMP 201, the combination showed a manageable safety profile, with only 10% treatment discontinuation due to adverse events.

- Common side effects:
  - Nausea (67%), Diarrhea (58%), Rash (50%) — mostly mild
  - CK elevation (~60%, grade ≥3 in 24%) — asymptomatic, managed with dose adjustments
  - Other mild AEs: Edema, fatigue, transaminase increase

Dose modifications (interruptions: ~80%, reductions: ~37%) were expected due to long treatment durations, but no new safety concerns emerged.

FDA Accelerated Approval (May 2025): Granted for KRAS-mutant recurrent LGSOC after prior therapy, marking one of the first targeted therapies for this disease. A Phase III confirmatory trial (RAMP 301) is ongoing to compare the combo against standard treatments, with PFS as the primary endpoint (65).

Non–Small Cell Lung Cancer (RAMP 202 and Other Trials)

The Phase II RAMP 202 trial evaluated avutometinib with or without defactinib in $KRAS^{G12V}$ -mutant NSCLC, a subgroup lacking targeted therapies. Results were disappointing—monotherapy showed a 0% response rate, and the combination yielded only an 11% ORR, with median progression-free survival around 4 months in both arms. Disease control rates were modest (37–44%) and below the threshold for further development. As a result, the trial was halted after Part A, and the combination was not advanced. Safety remained consistent with previous studies, with no new signals. Overall, the findings suggest that $KRAS^{G12V}$ NSCLC likely requires alternative strategies beyond FAK inhibition (66).

Recent efforts have focused on combining avutometinib with $KRAS^{G12C}$ inhibitors to improve outcomes in KRAS-mutant NSCLC. While $KRAS^{G12C}$ accounts for ~40% of KRAS-driven lung adenocarcinomas and agents like sotorasib and adagrasib show ~37–43% ORR as monotherapy, resistance often arises through pathway reactivation. Preclinical data support dual vertical inhibition—pairing a MEK clamp (avutometinib) with $KRAS^{G12C}$ blockade—to enhance ERK suppression and delay resistance. In the RAMP 203 trial, the avutometinib + sotorasib combo achieved a 40% ORR in $KRAS^{G12C}$-inhibitor–naïve patients, with deep responses. Importantly, it also showed 14% ORR in those previously resistant to $KRAS^{G12C}$ inhibitors, leading to an overall ORR of 25%. These encouraging results earned the combination a Fast Track designation from the FDA. The established RP2D is avutometinib 4 mg BID (21/28 days) + sotorasib 960 mg daily, with no major added toxicities or drug interactions. A separate trial, RAMP 204, is now evaluating avutometinib with adagrasib in patients who have progressed on prior G12C therapies. These combination approaches aim to deepen responses and prolong efficacy in $KRAS^{G12C}$-mutant NSCLC beyond what single-agent inhibitors can offer (67).

Table 4: Key Clinical Trials of Avutometinib (VS-6766) ± Combinations (2019–2025)

Trial (Phase)	Patient Population (Genotype)	Regimen	Notable Outcomes	Ref.
FRAME (Phase I)	Solid tumors with KRAS/NRAS/BRAF mutations (expansion cohorts in LGSOC, NSCLC, CRC, pancreatic, etc.)	Avutometinib 3.2 mg BID 2×/week + Defactinib 200 mg BID (3 wks on/1 off) – intermittent	– LGSOC (KRAS-mut): ORR 42%, mPFS 20.1 mo. – NSCLC (KRAS-mut): ORR ~11%, mPFS 3.5 mo. – CRC/Pancreatic: 0% ORR (no PR; mainly stable disease). – RP2D established; common AEs rash, CPK elevation (mostly grade 1–2).	(31)
RAMP 201 (Phase II)	Recurrent Low-Grade Serous Ovarian Cancer (LGSOC), stratified	Avutometinib 3.2 mg BID 2×/week + Defactinib	– Overall (n=109): ORR 31%, median DOR 31.1 mo, mPFS 12.9 mo. – KRAS-mutant: ORR 44%,	(65)

	by KRAS mutation	200 mg BID (3/1 schedule)	mPFS 22.0 mo. – KRAS–wild-type: ORR 17%, mPFS 12.8 mo. – Responses seen even after prior MEK inhibitor (45–60% ORR in combo arm vs 0% with monotherapy). – Accelerated FDA approval granted for KRAS-mut LGSOC (2025).	
RAMP 202 (Phase II)	KRAS^G12V-mutant NSCLC (previously treated)	Avutometinib 3.2 mg BID 2×/week ± Defactinib 200 mg BID (randomized)	– Monotherapy: ORR 0% (0/16 PR), DCR 44%. – +Defactinib combo: ORR 11% (2/19 PR), DCR 37%. – Limited efficacy; did not proceed to expansion (no "go" in adaptive design). – No new safety signals; combination not pursued further in G12V NSCLC.	(66)
RAMP 203 (Phase I/II)	KRAS^G12C-mutant NSCLC (± prior G12C inhibitor)	Avutometinib 4 mg BID 2×/week (21/28 days) + Sotorasib 960 mg QD	– KRAS G12C-inhibitor naïve: ORR ~40%. – KRAS G12C-inhibitor resistant: ORR ~14% (some responses despite prior failure). – Combined ORR ~25% (Fast Track designation by FDA). – RP2D established; well tolerated with no significant drug–drug interaction.	(67)
VS-6766 + Everolimus (Phase I)	Solid tumors with RAS mutations (dose escalation + NSCLC expansion)	Avutometinib (VS-6766) 3.2 mg BID 2×/week + Everolimus 5 mg 2×/week (intermittent)	– NSCLC (KRAS-mut): PRs in multiple KRAS variants; median PFS ~6.3 mo in refractory pts. – LGSOC (KRAS G12D): 2 confirmed PRs with extraordinarily long DOR (~36–42 mo ongoing). – Established tolerable dual-pathway blockade (MAPK + PI3K/mTOR); supports further trials in RAS-driven tumors.	(68)

Future Perspectives

The unique "RAF–MEK clamp" mechanism of avutometinib, coupled with its early clinical success, has important implications for future research and clinical development. One key priority is to harness avutometinib as a backbone for combination therapies to counteract compensatory pathways underlying resistance to MAPK inhibitors. Rational co-targeting approaches (e.g., combining avutometinib with FAK inhibitors, direct KRAS inhibitors, or EGFR/PI3K–mTOR pathway blockers) are expected to deepen responses and prolong disease control. Initial studies already demonstrate synergistic efficacy with such strategies: in RAS-driven ovarian cancer, avutometinib plus a FAK inhibitor achieved objective responses in nearly half of patients , and pairing it with a KRAS G12C inhibitor in lung cancer improved response rates even among patients with prior KRAS inhibitor resistance. These results underscore the importance of biomarkerdriven patient selection. For example, in KRAS-mutant low-grade serous ovarian cancer the ORR was ~44% versus ~17% in KRAS–wild-type patients, illustrating why patient selection by tumor genotype is crucial. In addition, expanding investigations into traditionally hard-to-treat RAS/MAPK-driven malignancies (e.g., KRAS-mutant pancreatic and colorectal cancers) is a critical next step, and combining avutometinib with standard treatments (such as chemotherapy or other targeted agents) in these settings could help overcome intrinsic resistance mechanisms and broaden its impact. Optimizing dosing – for instance, refining the intermittent schedule that maintains ERK suppression with lower toxicity – and integrating combinations earlier in treatment could further enhance efficacy and preempt resistance. By integrating these approaches (combination therapy, biomarker-driven patient selection, and proactive resistance mitigation), researchers can fully leverage avutometinib's novel mechanism to achieve more durable responses and improved outcomes across a broad spectrum of RAS/MAPK-driven cancers.

CONCLUSION

Avutometinib (VS-6766) represents a significant advancement in targeted cancer therapy through its novel "RAF–MEK clamp" mechanism, which addresses key limitations of classical MEK or RAF inhibitors. By stabilizing RAF–MEK complexes in an inactive state, avutometinib effectively suppresses ERK signaling while preventing feedback reactivation that commonly undermines MAPK pathway inhibition. Preclinical and clinical studies have consistently demonstrated its superior efficacy in RAS- and RAF-mutant malignancies, particularly in low-grade serous ovarian cancer and select subtypes of non–small cell lung cancer. Moreover, the drug's favorable tolerability profile, especially under intermittent dosing schedules, supports its long-term use in chronic treatment settings. The integration of avutometinib with rational combination strategies—such as focal adhesion kinase inhibitors or KRAS G12C inhibitors—has further enhanced its therapeutic impact, overcome resistance and expanding its utility across diverse tumor types. These promising findings not only validate the concept of dual RAF–MEK inhibition but also emphasize the need for continued clinical development, biomarker-guided therapy selection, and investigation of synergistic combinations. Taken together,

avutometinib emerges as a first-in-class targeted agent with the potential to reshape treatment paradigms for MAPK-driven cancers.

REFERENCES

1. Bahar ME, Kim HJ, Kim DR. Targeting the RAS/RAF/MAPK pathway for cancer therapy: from mechanism to clinical studies. Signal Transduction and Targeted Therapy. 2023;8(1):455.
2. Ullah R, Yin Q, Snell AH, Wan L. RAF-MEK-ERK pathway in cancer evolution and treatment. Semin Cancer Biol. 2022;85:123-54.
3. Guo YJ, Pan WW, Liu SB, Shen ZF, Xu Y, Hu LL. ERK/MAPK signalling pathway and tumorigenesis. Exp Ther Med. 2020;19(3):1997-2007.
4. Li Y, Dong Q, Cui Y. Synergistic inhibition of MEK and reciprocal feedback networks for targeted intervention in malignancy. Cancer Biol Med. 2019;16(3):415-34.
5. Song Y, Bi Z, Liu Y, Qin F, Wei Y, Wei X. Targeting RAS-RAF-MEK-ERK signaling pathway in human cancer: Current status in clinical trials. Genes Dis. 2023;10(1):76-88.
6. Singh, S., Virmani, R., & Virmani, T. G. (2016). Vitamin-B¬ 17: An alternative treatment of cancer-a myth or truth. *J. Mol. Pharmaceuticals Regulat. Affairs*, *1*, 1-5.
7. Isermann T, Sers C, Der CJ, Papke B. KRAS inhibitors: resistance drivers and combinatorial strategies. Trends in Cancer. 2025;11(2):91-116.
8. Yakubov R, Kaloti R, Persaud P, McCracken A, Zadeh G, Bunda S. It's all downstream from here: RTK/Raf/MEK/ERK pathway resistance mechanisms in glioblastoma. J Neurooncol. 2025;172(2):327-45.
9. Liu D, Xing M. Potent inhibition of thyroid cancer cells by the MEK inhibitor PD0325901 and its potentiation by suppression of the PI3K and NF-kappaB pathways. Thyroid. 2008;18(8):853-64.
10. Henderson Y, Chen Y, Frederick M, Lai S, Clayman G. MEK Inhibitor PD0325901 Significantly Reduces the Growth of Papillary Thyroid Carcinoma Cells In vitro and In vivo. Molecular Cancer Therapeutics. 2010;9:1968-76.
11. Sanchez JN, Wang T, Cohen MS. BRAF and MEK Inhibitors: Use and Resistance in BRAF-Mutated Cancers. Drugs. 2018;78(5):549-66.
12. Demirkiran C, Greenman M, Bellone S, McNamara B, Hartwich T, Manavella D, et al. Preclinical in vitro and in vivo activity of the RAF/MEK clamp avutometinib in combination with FAK inhibition in uterine carcinosarcomas. Gynecologic oncology. 2024;187:12-20.
13. McNamara B, Demirkiran C, Hartwich TMP, Bellone S, Manavella D, Mutlu L, et al. Preclinical efficacy of RAF/MEK clamp avutometinib in combination with FAK inhibition in low grade serous ovarian cancer. Gynecol Oncol. 2024;183:133-40.
14. Kumar, P., Mangla, B., Singh, S., & Rana, A. (2018). Drug master file: Global regulatory issues and challenges. European Journal of Biomedical and Pharmaceutical Sciences, 5(1), 623–626.

15. Stefanachi A, Leonetti F, Pisani L, Catto M, Carotti A. Coumarin: A natural, privileged and versatile scaffold for bioactive compounds. Molecules. 2018;23(2):250.
16. Singh S, Kumar P, Rana A. Global regulatory challenges of common technical document. World Journal of Pharmacy and Pharmaceutical Sciences. 2017;6(12), 1298-1309. DOI: 10.20959/wjpps201712-10616
17. Acharya KR, Sturrock ED, Riordan JF, Ehlers MR. Ace revisited: a new target for structure-based drug design. Nat Rev Drug Discov. 2003;2(11):891-902.
18. Aoki T, Hyohdoh I, Furuichi N, Ozawa S, Watanabe F, Matsushita M, et al. Optimizing the Physicochemical Properties of Raf/MEK Inhibitors by Nitrogen Scanning. ACS Med Chem Lett. 2014;5(4):309-14.
19. Mphahlele MJ, Onwu EE, Maluleka MM. Spectroscopic, X-ray Diffraction and Density Functional Theory Study of Intra- and Intermolecular Hydrogen Bonds in Ortho-(4-tolylsulfonamido)benzamides. Molecules. 2021;26(4).
20. Singh S, Gaikwad DD, Singh K. Formulation, development and investigation of matrix type sustained release tablet of antiulcer drug by using soluble polymer as a drug release retarding agent**.** International Journal of Membrane Science and Technology. 2023;10(4):2593–2603.
21. Ritt DA, Abreu-Blanco MT, Bindu L, Durrant DE, Zhou M, Specht SI, et al. Inhibition of Ras/Raf/MEK/ERK Pathway Signaling by a Stress-Induced Phospho-Regulatory Circuit. Mol Cell. 2016;64(5):875-87.
22. Shan KS, Rehman TU, Ivanov S, Domingo G, Raez LE. Molecular targeting of the BRAF proto-oncogene/mitogen-activated protein kinase (MAPK) pathway across cancers. International Journal of Molecular Sciences. 2024;25(1):624.
23. Li L, Zhao GD, Shi Z, Qi LL, Zhou LY, Fu ZX. The Ras/Raf/MEK/ERK signaling pathway and its role in the occurrence and development of HCC. Oncol Lett. 2016;12(5):3045-50.
24. Satbir Singh, Anjana Devi, Sonam Sharma, Sakshi Sabharwal, Shilpa Sharma, Simran Dhiman, Shreya Chauhan, A Review on Microspheres and Its Role in Different Drug Delivery System as a Novel Approach, Int. J. of Pharm. Sci., 2024, Vol 2, Issue 6, 1112-1126. https://doi.org/10.5281/zenodo.12507394
25. Gonzalez-Del Pino GL, Li K, Park E, Schmoker AM, Ha BH, Eck MJ. Allosteric MEK inhibitors act on BRAF/MEK complexes to block MEK activation. Proc Natl Acad Sci U S A. 2021;118(36).
26. Oh YT, Deng J, Yue P, Sun SY. Paradoxical activation of MEK/ERK signaling induced by B-Raf inhibition enhances DR5 expression and DR5 activation-induced apoptosis in Ras-mutant cancer cells. Sci Rep. 2016;6:26803.
27. McCubrey JA, Steelman LS, Chappell WH, Abrams SL, Wong EW, Chang F, et al. Roles of the Raf/MEK/ERK pathway in cell growth, malignant transformation and drug resistance. Biochim Biophys Acta. 2007;1773(8):1263-84.
28. Ishii N, Harada N, Joseph EW, Ohara K, Miura T, Sakamoto H, et al. Enhanced inhibition of ERK signaling by a novel allosteric MEK inhibitor, CH5126766, that suppresses feedback reactivation of RAF activity. Cancer Res. 2013;73(13):4050-60.

29. Edwards A, Stalnecker C, Morales A, Taylor K, Klomp J, Klomp J, et al. TEAD Inhibition Overcomes YAP1/TAZ-Driven Primary and Acquired Resistance to KRAS Inhibitors. Cancer Research. 2023;83:4112-2
30. Marasco M, Kumar D, Seale T, Borrego SG, Kaplun E, Aricescu I, et al. Concurrent SOS1 and MEK suppression inhibits signaling and growth of NF1-null melanoma. Cell Reports Medicine. 2024;5(11):101818.
31. Banerjee S, Krebs MG, Greystoke A, Garces AI, Perez VS, Terbuch A, et al. Defactinib with avutometinib in patients with solid tumors: the phase 1 FRAME trial. Nature Medicine. 2025.
32. Wada M, Horinaka M, Yamazaki T, Katoh N, Sakai T. The dual RAF/MEK inhibitor CH5126766/RO5126766 may be a potential therapy for RAS-mutated tumor cells. PloS one. 2014;9(11):e113217.
33. Huang W, Yang AH, Matsumoto D, Collette W, Marroquin L, Ko M, et al. PD0325901, a mitogen-activated protein kinase kinase inhibitor, produces ocular toxicity in a rabbit animal model of retinal vein occlusion. J Ocul Pharmacol Ther. 2009;25(6):519-30.
34. Wu LW, Jang SJ, Shapiro C, Fazlollahi L, Wang TC, Ryeom SW, et al. Diffuse Gastric Cancer: A Comprehensive Review of Molecular Features and Emerging Therapeutics. Target Oncol. 2024;19(6):845-65.
35. Coma S, Paradis J, Gutkind JS, Pachter J. Abstract 1425: Synergistic antitumor efficacy of the dual RAF/MEK inhibitor VS-6766 with FAK inhibition for treatment of RAS-dependent solid tumors2021. 1425- p.
36. Singh K, Gaikwad DD, Singh S. Formulation design and development of fast disintegrating tablets of losartan potassium by using locust bean gum as superdisintegrant. International Journal of Membrane Science and Technology, 2023, Vol. 10, No. 5, pp 1084-1096.
37. Leicht DT, Balan V, Zhu J, Kaplun A, Bronisz A, Rana A, et al. MEK-1 activates C-Raf through a Ras-independent mechanism. Biochim Biophys Acta. 2013;1833(5):976-86.
38. Rosell R, Codony-Servat J, González J, Santarpia M, Jain A, Shivamallu C, et al. KRAS G12C-mutant driven non-small cell lung cancer (NSCLC). Critical Reviews in Oncology/Hematology. 2024;195:104228.
39. Alkarain A, Slingerland J. Deregulation of p27 by oncogenic signaling and its prognostic significance in breast cancer. Breast Cancer Res. 2004;6(1):13-21.
40. Nheu T, He H, Hirokawa Y, Walker F, Wood J, Maruta H. PAK is essential for RAS-induced upregulation of cyclin D1 during the G1 to S transition. Cell Cycle. 2004;3(1):71-4.
41. Satbir Singh, Kehar Singh, Yogita, Hemant Rana, A Review On Gastro Retentive Drug Delivery System: Novel Approach With The Future Perspectives , Int. J. of Pharm. Sci., 2024, Vol 2, Issue 6, 606-621. https://doi.org/10.5281/zenodo.11550881
42. Grisham R, Monk B, Nieuwenhuysen E, Moore K, Fabbro M, O'Malley D, et al. GOG-3097/ENGOT-ov81/GTG-UK/RAMP 301: a phase 3, randomized trial evaluating avutometinib plus defactinib compared with investigator's choice of treatment in patients with recurrent low grade serous ovarian cancer. International Journal of Gynecologic Cancer. 2024.

43. Hu HH, Wang SQ, Shang HL, Lv HF, Chen BB, Gao SG, et al. Roles and inhibitors of FAK in cancer: current advances and future directions. Front Pharmacol. 2024;15:1274209.
44. Kumar P, Mangla B, Singh S. Pitavastatin: a potent drug. *Int J Pharma Res Health Sci*. 2018;6(1):2070–2074. DOI:10.21276/ijprhs.2018.01.01
45. Adderley H, Blackhall FH, Lindsay CR. KRAS-mutant non-small cell lung cancer: Converging small molecules and immune checkpoint inhibition. EBioMedicine. 2019;41:711-6.
46. Zhu C, Guan X, Zhang X, Luan X, Song Z, Cheng X, et al. Targeting KRAS mutant cancers: from druggable therapy to drug resistance. Mol Cancer. 2022;21(1):159.
47. Indini A, Rijavec E, Ghidini M, Cortellini A, Grossi F. Targeting KRAS in Solid Tumors: Current Challenges and Future Opportunities of Novel KRAS Inhibitors. Pharmaceutics. 2021;13(5).
48. Singh S, Mittal H, Singh K, Rana H. A review on chemistry, synthesis, extraction, nano-formulation and therapeutic use of curcumin. Asian Journal of Research in Pharmaceutical Sciences. 2025;15(2):135–140.
49. Zhou K, Liu Y, Tang C, Zhu H. Pancreatic Cancer: Pathogenesis and Clinical Studies. MedComm (2020). 2025;6(4):e70162.
50. Singhal A, Li BT, O'Reilly EM. Targeting KRAS in cancer. Nat Med. 2024;30(4):969-83.
51. Melosky B, Juergens RA, Banerji S, Sacher A, Wheatley-Price P, Snow S, et al. The continually evolving landscape of novel therapies in oncogene-driven advanced non-small-cell lung cancer. Ther Adv Med Oncol. 2025;17:17588359241308784.
52. Yoshimura A, Horinaka M, Yaoi T, Ono H, Itoh K, Yamada T, et al. Epithelial-mesenchymal transition status is a remarkable biomarker for the combination treatment with avutometinib and defactinib in KRAS-mutated non-small cell lung cancer. Br J Cancer. 2024;131(2):361-71.
53. Hartwich TMP, Mansolf M, Demirkiran C, Greenman M, Bellone S, McNamara B, et al. Preclinical evaluation of avutometinib and defactinib in high-grade endometrioid endometrial cancer. Cancer Med. 2024;13(17):e70210.
54. Brastianos P, Twohy E, Gerstner E, Kaufmann T, Iafrate A, Lennerz J, et al. Alliance A071401: Phase II Trial of Focal Adhesion Kinase Inhibition in Meningiomas With Somatic NF2 Mutations. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2022;41:JCO2102371.
55. Grisham R, Aghajanian C, Van Nieuwenhuysen Els, Rodrigues M, Ring K, Santin A, et al. PO004LBA/#1515 Efficacy and safety of avutometinib + defactinib in recurrent low-grade serous ovarian cancer following prior systemic therapy. International Journal of Gynecological Cancer. 2023;33:A3-A4.
56. Lubrano S, Cervantes-Villagrana RD, Faraji F, Ramirez S, Sato K, Adame-Garcia SR, et al. FAK inhibition combined with the RAF-MEK clamp avutometinib overcomes resistance to targeted and immune therapies in BRAF V600E melanoma. Cancer Cell. 2025;43(3):428-45.e6.

57. Almazan J, Turapov T, Kircher DA, Stanley KA, Culver K, Medellin AP, et al. Combined inhibition of focal adhesion kinase and RAF/MEK elicits synergistic inhibition of melanoma growth and reduces metastases. Cell Rep Med. 2025;6(2):101943.
58. Weigelt B, Warne PH, Lambros MB, Reis-Filho JS, Downward J. PI3K pathway dependencies in endometrioid endometrial cancer cell lines. Clin Cancer Res. 2013;19(13):3533-44.
59. Kakadia S, Yarlagadda N, Awad R, Kundranda M, Niu J, Naraev B, et al. Mechanisms of resistance to BRAF and MEK inhibitors and clinical update of US Food and Drug Administration-approved targeted therapy in advanced melanoma. Onco Targets Ther. 2018;11:7095-107.
60. Guo C, Banerji U. Searching for treatments for non-G12C-KRAS mutant cancers. British Journal of Cancer. 2021;125.
61. Frisch A, Martin E, Kim SY, Riess JW, Sen T, Karim N. KRAS mutated NSCLC: past, present, and future directions in a rapidly evolving landscape. Oncologist. 2025;30(6).
62. Corcoran RB, Do KT, Kim JE, Cleary JM, Parikh AR, Yeku OO, et al. Phase I/II Study of Combined BCL-xL and MEK Inhibition with Navitoclax and Trametinib in KRAS or NRAS Mutant Advanced Solid Tumors. Clin Cancer Res. 2024;30(9):1739-49.
63. Salgia R, Pharaon R, Mambetsariev I, Nam A, Sattler M. The improbable targeted therapy: KRAS as an emerging target in non-small cell lung cancer (NSCLC). Cell Rep Med. 2021;2(1):100186.
64. Perrone C, Angioli R, Luvero D, Giannini A, Di Donato V, Cuccu I, et al. Targeting BRAF pathway in low-grade serous ovarian cancer. J Gynecol Oncol. 2024;35(4):e104.
65. Avutometinib Plus Defactinib Gains FDA Approval in KRAS+ Ovarian Cancer [Internet]. 2025. Available from: https://www.targetedonc.com/view/avutometinib-plus-defactinib-gains-fda-approval-in-kras-ovarian-cancer.
66. Reuss JE, Gandhi SG, Spigel DR, Janne PA, Paz-Ares LG, Gadgeel SM, et al. RAMP 202: A phase 2 study of avutometinib (VS-6766)±defactinib, in patients with advanced KRAS G12V mutant non–small cell lung cancer (NSCLC). J Clin Oncol. 2023;41(Suppl 16):9100.
67. FDA Grants Avutometinib/Sotorasib FTD in KRAS-Mutated NSCLC [Internet]. 2024. Available from: https://www.targetedonc.com/view/fda-grants-avutometinib-sotorasib-ftd-in-kras-mutated-nsclc.
68. Minchom A, Denis L, Spicer J. Phase I trial of the RAF/MEK clamp VS-6766 in combination with everolimus using intermittent dosing schedule with expansion in NSCLC across multiple KRAS variants.