Background

The transmission of extracellular signals into intracellular responses is a complex process which often involves the activity of one or more mitogen-activated protein kinases (MAPKs). The activation of a MAPK employs a core three-kinase cascade consisting of a MAPK kinase kinase (MAP3K or MAPKKK) which phosphorylates and activates a MAPK kinase (MAP2K, MEK, or M KK) which then phosphorylates and increases the activity of one or more MAPKs. Upon activation, MAPKs can phosphorylate a variety of intracellular targets including transcription factors, nuclear pore proteins, membrane transporters, cytoskeletal elements, and other protein kinases.

Discovery of MAPKs

The MAPKs extracellular signal regulated protein kinases 1 and 2 (ERK1/2) were first identified as mitogen-stimulated ~42 kDa phosphoproteins in the early 1980s, and later as insulin and nerve growth factor (NGF)-stimulated activities that retained the ability to phosphorylate the model substrates microtubule-associated protein-2 (MAP2) and myelin basic protein (MBP).

In the following years, the MAPK family was discovered to include three c-Jun N-terminal kinases (JNK), four p38 isoforms, ERK3 isoforms, ERK5 and ERK7. The first JNK family members were independently identified as cycloheximide-activated MBP kinases and purified due to their ability to interact with the N-terminus of the transcription factor c-Jun. PCR-based cloning strategies and a two-hybrid screen led to the discovery of additional JNK and p38 isoforms and ERKs 5 and 7 reviewed by Chen et al. A summary of the cellular processes involving

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these MAPKs is shown in Figure 2. Detailed reviews by Adnane et al., Daaka et al and Dang et al. are also recommended for further information.1,16,17

Upstream regulation of ERK1/2

The collaborative findings from a number of laboratories led to the connection of ERK1/2 to their upstream regulators MAPK/ERK kinase 1 and 2 (MEK1/2); the identification of Raf isoforms as upstream activators of these MAP2Ks; and the observation that Raf is an effector of Ras small GTP binding proteins.18-24 Isoforms of Ras and Raf are found mutated in a variety of human tumors, implicating ERK1/2 in proliferation and oncogenic growth.25,26 Although regulation through this pathway is exceedingly complex, the potential of this MAPK pathway to promote tumorigenesis was later supported by the demonstration that an activated mutant of MEK1 transformed cells and promoted growth of tumors in nude mice.27 Subsequently, through the use of dominant interfering mutants, pharmacological inhibitors of MEK1/2, gene disruption and RNA interference, these ubiquitous kinases have been shown to be intimately involved in normal processes including embryogenesis, cell differentiation, glucose sensing and synaptic plasticity.28-33

MAP2Ks

MEK1, an exemplary MAP2K, was purified as a ~45 kDa biological activator of ERK1/2.34-36 The identification of additional MAP2Ks (MEKs 2-7), all of roughly equivalent size, employed DNA-based molecular techniques as opposed to protein purification reviewed by Chen et al.15 and Lewis et al.16 These kinases are unusual in that they are dual-specificity kinases, phosphorylating both tyrosine and serine/threonine residues. Unlike MAPKs, which phosphorylate a wide range of proteins, MAP2Ks are highly specific: they are dedicated to phosphorylation of only one or a couple of MAPKs and few, if any, other substrates. MAP2Ks integrate signals from multiple regulatory inputs and serve as points of signal integration, in part through scaffolding proteins and docking site-mediated protein-protein interactions.

Raf isoforms and other MAP3Ks

The most readily identifiable feature of MAPK signaling is the three kinase cascade consisting of a MAP3K, a MAP2K and a MAPK. The three-kinase organization of this cascade is identical to that of the three-kinase cascade of Ste11 (MAP3K) - Ste7 (MAP2K) - Fus3/Kss1 (MAPK) in the yeast pheromone response pathway.37 MAP2Ks and MAPKs are related in sequence throughout metazoans, although Raf proteins do not seem to have counterparts identified in yeast. Interestingly, Raf was originally discovered as a retroviral oncogene.38 Three isoforms, c-Raf (or Raf-1), B-Raf and A-Raf, are found in mammals. In addition to the core ~35 kDa kinase domain, Raf proteins contain an N-terminal regulatory region, also about 35-40 kDa, which can bind Ras. Raf proteins specifically phosphorylate only the MAP2Ks MEKs1 and 2, and were initially thought to function in a tissue-specific manner. More recent studies, aided by the development of B-Raf inhibitors as anticancer agents, led to the understanding that Raf isoforms dimerize.39,40 The unanticipated actions of these B-Raf inhibitors provoked more in-depth molecular analysis showing that dimerization can enhance Raf activity and that Raf heterodimers have different activities.41-43 Two other enzymes that function as MAP3Ks in the ERK1/2 pathway are Mos and Tpl2 (Cot), both originally identified as proteins that could transform cells.44,45 These enzymes function only in specialized situations and when present, they activate the cascade; Mos is expressed primarily in oocytes, while Tpl2 is stabilized in response to lipopolysaccharide.46,47

The parallels between yeast and mammalian signaling led investigators to search for Ste11 homologs in mammals. MEKK1 was the first mammalian MAP3K identified from its homology to Ste11.48,49 In contrast to the selectivity displayed by Raf MAP3Ks, MEKK1, a large protein of 195 kDa, displayed the ability to phosphorylate several MAP2Ks (MEKs 1-4, 6 and 7) in vitro. Early evidence suggested that MEKK1 was a regulator of MEK1/2, but gene disruption experiments and numerous biochemical analyses indicate that MEKK1

Figure 1 | MAP3K and MAP2K compounds

![MAP3K and MAP2K compounds](image-url)
predominantly coordinates downstream signaling through activation of MEKs 4 and 7 and the JNK pathway.\textsuperscript{50-53} Subsequent isolation of related cDNAs have led to the discovery of a family of related enzymes (i.e. MEKks 1-4; ASK1,2; TAK1) reviewed by Raman et al\textsuperscript{54} and Johnson et al.\textsuperscript{54} As a group, these enzymes also display broader substrate specificity than Raf and probably regulate multiple MAPK pathways in context-dependent processes. Non-Ste11 homologs, such as the Ste20 homologs TAOs 1-3 are MAP3Ks that regulate the p38 pathway.\textsuperscript{1}

Figure 2 presents a simplified model of the organization of MAPK cascades. Of note are the number of MAP2K-MAPK combinations a given MAP3K can regulate and the resulting points of cross-talk. How the organization of MAPK cascades affects their function will be discussed next.

Properties of MAPK cascades
Signaling features of both mammalian Raf-MEK1/2-ERK1/2 and the yeast cascades initially provided an insight into the primary features of MAPK signaling. Generally similar mechanisms of regulation exist in other MAPK cascades, although it is expected that additional novel features will be found. The impact of scaffolds and cascade localization are still not well integrated into current models. As mechanistic insight has accumulated, the complexity of these pathways, in spite of the apparent simplicity of the three-kinase unit, must be acknowledged. Also the regulatory plasticity that accrues from the three-kinase cascade should not be underestimated.

The conventional view of a signaling cascade was developed from the first studied kinase pathway, cAMP-dependent protein kinase (PKA). Amplification occurred because components were more abundant moving down the cascade. The cooperative, switch-like behavior in this pathway derives from the requirement of four cAMP molecules to activate PKA.\textsuperscript{55} This requirement may not apply unilaterally to MAPK pathways. MEK1/2 are much more abundant than Raf proteins, but MEK1/2 are in some cases as abundant as ERK1/2.\textsuperscript{56,57} The MAPK pathway exhibits a similar switch-like behavior that guarantees a threshold to prevent activation of the pathway by noise in the system. This behavior is mechanistically distinct and partly derives from nonprocessive dual phosphorylation of MAPKs by MAP2Ks. MAPKs contain a poorly conserved loop that lies C-terminal to the catalytic residues, referred to as the activation loop. This loop contains a TXY motif. Phosphorylation of both the threonine and tyrosine residues of this motif by MAP2Ks is required to activate MAPKs.\textsuperscript{58} In cells, the phosphorylation of tyrosine before threonine introduces the activation threshold and rapid cooperative activation.\textsuperscript{59,60}

Figure 2 | MAP Kinase Networks
The existence of three proteins in series provides for multiple points of regulatory input. For example, Raf isoforms are phosphorylated on numerous sites by several protein kinases that increase or decrease activity and influence protein-protein interactions.\(^6\) Prominent among these regulatory inputs are sites of feedback phosphorylation by ERK1/2 that interfere with re-activation of Raf by Ras.\(^5,6\) Raf isoforms also interact with a variety of adaptor proteins.\(^6\) MAP2Ks are also phosphorylated at other sites in addition to activating sites in their activation loops.\(^5,6,7\) For example, phosphorylation of MEK1/2 in unique insert regions disrupts their ability to interact with Raf.\(^5,6,8,9\)

To summarize, the fidelity of signaling to ERK1/2 is dictated by the integration of a broad collection of signals that can be communicated at multiple levels in the pathway.

**Scaffolding proteins**

Scaffolds have a major influence on cascade function strongly impacting on the activities and outputs of MAPK pathways. Best described by the yeast example Ste5, scaffolds are paramount for achieving MAPK specificity.\(^5,7\) The yeast MAP3K, Ste11, can activate either Ste7, a MAP2K for the MAPKs Kss1 and Fus3, in response to pheromone; or Pbs2, the MAP2K for Hog1 (the yeast p38 MAPK) in response to osmotic stress.\(^7\) Scaffolding proteins dictate which signal activates Ste11 and which MAP2K is targeted for activation by Ste11. In the pheromone response, Ste5 scaffolds the interaction of Ste11 with Ste7, whereas the ability of Pbs2 to create a stable interaction of Ste11 with the osmosensor Sho1 allows Ste11 to act in the osmotic response.\(^7\)

As noted above, mammalian signaling has similar complexities, suggested by the many observations showing that individual MAP3Ks can regulate multiple MAPK cascades. Nevertheless, no obvious Ste5 homolog has been identified in mammals. The scaffolding work is likely to be distributed among several different types of proteins in mammals and may often be “wrapped” within some of the upstream enzymes. Several MAP3Ks contain docking sites that allow them to bind stably to specific MAPKs. For example, MEKK1 binds tightly to JNKs through a docking motif.\(^7\) Stable interactions with MAPKs may be mediated by at least two short sequence motifs, the docking (D or DEJL) motif and the FXF (DEF) motif.\(^5,7\) One or more of these motifs are often present in scaffolds, activators, certain phosphatases and many substrates. JNK-interacting protein (JIP) scaffolds which organize JIP and sometimes p38 MAPK pathways have some functional parallels to Ste5s.\(^5,7\) The scaffold kinase suppressor of Ras (KSR) was discovered in the sevenless eye development pathway in *Drosophila* and in the vulval induction pathway of *C. elegans*.\(^7,8\) KSR proteins are members of the Raf family, but are pseudokinases because they lack the essential ATP-binding lysine residue. In mammals KSR1 and KSR2 bind ERK1/2, MEK1/2 and Raf isoforms. They interact with Raf proteins and can allosterically activate Raf.\(^4\) Other proteins that influence assembly and activation of MAPK pathways include Sur8, CNK, MP-1 and IMP.\(^6,7\)

**Activation of MAPKs from the cell surface**

**Tyrosine kinase receptor activation of ERK1/2**

ERK1/2 are activated by a wide variety of stimuli that act through cell surface receptors. Of all the signaling pathways emanating from these receptors, the pathway from receptor tyrosine kinases to ERK1/2 is the best delineated.\(^1,16,17\) Ligand binding to receptor tyrosine kinases stimulates homo- and/or heterodimerization of the receptors and increases their tyrosine kinase activity. Activated receptors can then phosphorylate themselves and their dimerization partners, creating phosphotyrosines motifs. These motifs are recognized by SH2 domains that exist in a variety of proteins including the adaptor proteins Shc and Grb2. The SH3 domain of the Ras guanine nucleotide exchange factor son of sevenless (SOS) interacts with proline-rich regions on the receptor-bound adaptor proteins, completing the formation of a Ras-activating complex at the plasma membrane. After association with the receptor-adaptor protein complex, SOS stimulates the exchange of GDP for GTP on Ras. When GTP-bound, Ras interacts with a number of downstream effectors including Raf.\(^20,21\)

The direct interaction of Ras with Raf isoforms localizes them to the plasma membrane, which may serve to bring them in proximity to non-receptor kinases such as Src family members, and serine/threonine kinases including p21-activated kinases (PAKs) and protein kinase C (PKC) isoforms. These kinases may phosphorylate Ras-Raf isoforms and further increase their activity towards substrates or enhance their interactions with other proteins.\(^8,17\) Upon activation, Raf isoforms can activate the MEK1/2-ERK1/2 pathway as discussed above.

**Activation of MAPKs by G-protein-coupled receptors**

Many hormones act through G-protein-coupled receptors (GPCRs) to increase ERK1/2 activity. Agonist binding to Gαs-coupled receptors results in the activation of adenyl cyclase, which raises the intracellular concentration of cAMP. Elevated cAMP levels can increase, decrease or have no effect on the activity state of ERK1/2 in a manner dependent on cell type, and perhaps other factors yet to be defined.\(^1\) Inhibition of ERK1/2 activity is believed to involve phosphorylation of serines 43 and 621 on
c-Raf by cAMP-activated protein kinase (PKA). The N-terminal phosphorylation is reported to reduce the ability of c-Raf to interact with Ras. Phosphorylation of serine 621 reduces c-Raf activity by disturbing binding to 14.3.3 protein.

Increasing cAMP levels in cells of neuroendocrine origin typically stimulates ERK1/2 activity. The activation of ERK1/2 may involve Ras itself or the Ras-related monomeric G-protein Rap-1, which can be activated by cAMP-activated guanine nucleotide exchange factors (GEFs). It has also been suggested that Gαs coupled receptor activation of ERK1/2 involves Src. Src may be activated by direct interaction with β-arrestin molecules that are engaged with internalized GPCRs. In either case, activated Src can direct influence c-Raf activity by phosphorylating sites that lie N-terminal to its kinase domain or by stimulating recruitment of SOS to receptor tyrosine kinases (RTKs).

In vivo evidence indicates Gαi-coupled receptor activation of ERK1/2 most likely employs the βγ subunits of heterotrimeric G-protein complexes. Through use of a βγ sequestering peptide, it has been suggested βγ subunits are necessary for Gαi-coupled receptor ligand stimulation of ERK1/2. Consistent with this observation, exogenous expression of βγ is sufficient to activate ERK1/2. Both Gαi-coupled receptor ligands and overexpressed βγ subunits require Src activity to stimulate ERK1/2. Src activation by βγ subunits is not likely to involve direct interaction of the proteins; rather it has been proposed that PI-3 kinase γ serves as an intermediary in the activation mechanism. Regardless of the mechanism employed, Src can activate ERK1/2 in the same manner as described for Src activation of ERK1/2 stimulated by Gαs.

Activation of ERK1/2 by Gαq-coupled receptors may require both Ras and PKC. Upon activation, Gαq directly activates PLCβ, which cleaves phosphatidylinositol 4,5-bisphosphate (PIP2) to generate inositol triphosphate (IP3) and diacylglycerol (DAG). The DAG produced and the increase in intracellular Ca2+ resulting from IP3 production can activate certain PKC isofoms. Phosphorylation of c-Raf by PKC can increase c-Raf activity.

**Inhibition of MAPK pathways**

**MAPK function studied by inhibition**

Researchers utilize loss-of-function experiments such as dominant-negative mutants, gene silencing by RNA interference, and inhibitors of components of the MAPK signaling pathway to determine the dependence of a particular cellular function on a kinase pathway. Both a troublesome and a useful feature to the dominant-negative approach is that MAPKs are activated by overlapping upstream pathways and share common substrates causing the dominant-negative mutant to inhibit more than one target. Thus, a kinase-dead MAP2K may inhibit the activation of all the MAPKs it regulates. Gene silencing approaches using RNA interference may require targeting multiple closely related enzymes and rescue experiments are important to demonstrate that a phenotype is not due to off-target effects. Pharmacological inhibitors of components of the MAPK pathway are often a viable alternative or a complementary tool in understanding the functional requirement in a given pathway. Many inhibitors bind to the ATP binding pocket common to all protein kinases. Allosteric inhibitors are becoming increasingly valuable to achieve greater specificity. The specificity derived from interacting outside the ATP pocket has continued to spur the search for inhibitors of many kinases that bind to other pockets on kinase surfaces. Some inhibitors are currently being developed that block essential protein-protein interactions.

**Inhibitors of the ERK1/2 pathway**

Selective ERK1/2 inhibitors have only recently been described. One such inhibitor, FR180204, competes for the ATP-binding pocket of ERK2, with IC50 values of 0.14 and 0.31 µM for ERK2 and ERK1 respectively. The utility of this compound is not yet certain. In the mid 1990’s the MEK1/2 inhibitors PD 98059 and U0126, used to interfere with the

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**Figure 3 | JNK and ERK compounds**

<table>
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<tr>
<th>BI 78D3 (3314)</th>
<th>SP 600125 (1496)</th>
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<td>Selective, competitive JNK inhibitor</td>
<td>Novel and selective JNK inhibitor</td>
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<th>FR 180204 (3706)</th>
<th>XMD 8-92 (4132)</th>
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<tr>
<td>Selective ERK inhibitor</td>
<td>ERK5 inhibitor</td>
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www.tocris.com | Functions and Modulation of MAP Kinase Pathways
ERK1/2 pathway, became available. PD 98059 was found in an in vitro kinase activation assay, while U0126 was identified in a cell-based assay as an inhibitor of AP-1 transcriptional activity. In contrast to the usual ATP site inhibitors, U0126 and PD 98059 bind outside of the ATP binding site, selecting a low activity conformation and thereby keeping or shifting the protein to the inactive state. Due to their mode of binding, these drugs are among the most selective inhibitors available. The only other kinase affected by these drugs is the related MAP2K MEK5, which is inhibited at only slightly higher concentrations than MEK1/2. Both drugs are useful at low micromolar concentrations and inhibit activation of MEK1/2, but require higher concentrations to block already activated MEK1/2 in cells. Subsequently, a number of MEK1/2 inhibitors have been developed that are much more potent and have variable specificity relative to MEK5, including PD 198306 and PD 0325901. PD 0325901 inhibits ERK1/2 activation in cells at concentrations as low as 25 nM, but fails to inhibit a large panel of other protein kinases at more than 100 times the concentration. ERK5 activation was inhibited at low micromolar concentrations, consistent with a ~10-fold greater potency towards MEK1/2 than MEK5. ARY-142886 (AZD6244) is another potent, noncompetitive inhibitor of MEK1/2 with a reported IC50 of 14 nM against purified MEK1.

Another expanding array of compounds to block the ERK1/2 pathway is directed against Raf isoforms. B-Raf has been an attractive target particularly because a B-Raf mutant, V600E, is found in a large percentage of melanomas. The Raf inhibitors, overall, are less selective than MEK1/2-directed drugs and include GW 5074, ZM 336372, and BAY 43-9006 (Sorafenib). BAY 43-9006 inhibits c-Raf and B-Raf with IC50 values in the nanomolar range, but also has significant activity towards several receptor tyrosine kinases.

Inhibitors of JNK pathways
SP 600125 is the most frequently used inhibitor of the JNK signaling pathway and blocks JNKs at concentrations in the range of 50-100 nM. SP 600125 inhibits several other protein kinases with roughly equal potency. CEP-1347 (KT-7515) blocks the JNK pathway but is specifically an inhibitor of the upstream mixed lineage kinases (MLKs). Thus, it will not block JNKs that are regulated in an MLK-independent manner.

Inhibitors of p38 signaling pathway
p38 inhibitors including SB 203580 have been developed using pyridinyl imidazoles as lead compounds. Crystallography showed that SB 203580 binds to the active site of p38, which prevents ATP binding. SB 203580 and several related compounds inhibit p38α and β, but not p38δ and γ. Structural studies of p38 identified a key feature of the ATP binding pocket that impacts inhibitor specificity. The threonine 106 residue of p38α and β is often larger than others, such as methionine or glutamine present in protein kinases such as p38δ and γ. This residue is called the gatekeeper. Enzymes with small gatekeeper residues can accommodate larger compounds in their ATP sites. As a consequence, Raf isoforms (due to their small gatekeeper side chains) can interact with p38 inhibitors, while p38δ and γ cannot. BIRB796, a diaryl urea compound, structurally unrelated to SB 203580, inhibits all four p38 isoforms by indirectly competing with the binding of ATP. BIRB796 binding requires a large conformational change in a conserved catalytic loop (DFG motif) of p38. This remodeled structure is unable to bind ATP.

Future prospects
At present we have an ample amount of information on components involved in the MAPK signaling pathway. Likewise, many substrates have been identified and mapped to functions in specific processes. Unfortunately, our understanding of these pathways is unilateral and often excludes feedback mechanisms, spatio-temporal aspects and context-specific signaling. Finally, uncovering how the MAPK pathway regulates, or is regulated by, newly discovered processes is an exciting task. Increasing amounts of evidence points to a role for MAPK in disease. p38 MAPK and JNK are potential

Figure 4 | p38 MAPK compounds

SB 203580 (1402) Selective inhibitor of p38 MAPK
SB 239063 (1962) Potent, selective p38 MAPK inhibitor; orally active
SX 011 (3639) p38 MAPK inhibitor
VX 745 (3915) Potent and selective p38α inhibitor
targets for drug development in neuronal disease as their inhibition may reduce the production of inflammatory cytokines known to be involved in a number of neural diseases such as cerebral ischemia, Alzheimer’s disease and Parkinson's disease.\textsuperscript{121} Mutations in signaling components that activate ERK have been found in many forms of cancer. Specifically, mutations in K-Ras are prominent in colon and pancreatic cancer; N-Ras mutations occur in melanomas; H-Ras mutations in cervical and bladder cancer; while B-Raf mutations are found in over 65\% of malignant melanomas.\textsuperscript{122,123} The ERK signaling pathway is a main component in several steps of tumorigenesis including cancer cell proliferation, migration, invasion and survival. A deeper understanding of MAPK signaling pathways is required for the development of new therapeutic drugs for various disease states.
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MAPK Compounds Available from Tocris

MAP3K- MAP2K- MAPK module

**ERK**
- GW 5074: Potent, selective c-Raf1 kinase inhibitor
- L-779,450: Potent Raf kinase inhibitor
- (5Z)-7-Oxoeaenol: Potent and selective TAK1 MAPKKK inhibitor
- ZM 336372: Potent, selective c-Raf inhibitor

**MAPK**
- Arctigenin: Potent MKK1 inhibitor. Also inhibits IκBα phosphorylation
- PD 98059: Specific inhibitor of MEK
- PD 198306: Selective inhibitor of MEK1/2
- SL 327: Selective inhibitor of MEK1 and MEK2; brain penetrant
- U0124: Inactive analog of U0126 (Cat. No. 1144)
- U0126: Potent, selective inhibitor of MEK1 and 2

**JNK**
- AEG 3482: Inhibitor of JNK signaling
- BI 78D3: Selective, competitive JNK inhibitor
- JIP-1 (153-163): JNK-selective inhibitor peptide
- SP 600125: Novel and selective JNK inhibitor
- SU 3327: Selective JNK inhibitor
- TCS JNK 5a: Selective inhibitor of JNK2 and JNK3

**p38**
- Anisomycin: Activates JNK/SAPK/p38 MAPK
- CMPD-1: Non-ATP-competitive p38α inhibitor
- JX 401: Potent, reversible p38 inhibitor
- RWJ 67657: Potent, selective p38α and p38β inhibitor
- SB 202190: Selective inhibitor of p38 MAPK
- SB 203580: Selective inhibitor of p38 MAPK
- SU 3327: Selective JNK inhibitor
- TCS JNK 5a: Selective inhibitor of JNK2 and JNK3

Receptor and Upstream Signaling

**FAK**
- AG 490: EGFR-kinase inhibitor. Also JAK2, JAK3 inhibitor
- AG 1478 hydrochloride: Highly potent EGFR-kinase inhibitor
- BIBX 1382 dihydrochloride: Highly selective EGFR-kinase inhibitor
- Iressa: Orally active, selective EGFR inhibitor

**RTKs**

**EGFR**
- AG 490: EGFR-kinase inhibitor. Also JAK2, JAK3 inhibitor
- AG 1478 hydrochloride: Highly potent EGFR-kinase inhibitor
- BIBX 1382 dihydrochloride: Highly selective EGFR-kinase inhibitor
- Iressa: Orally active, selective EGFR inhibitor

**VEGFR/PDGFR**
- Demethylasterriquinone B1: Selective insulin RTK activator
- K 252a: Protein kinase inhibitor
- PHA 665752: Potent and selective MET inhibitor
- Pircropodophyllotoxin: Selective IGF1R inhibitor
- SU 16f: Potent and selective PDGFRβ inhibitor
- SU 4312: Potent inhibitor of VEGFR tyrosine kinase
- SU 5402: Potent FGFR and VEGF inhibitor
- SU 5416: VEGF inhibitor. Also inhibits KIT, RET, MET and FLT
- ZM 323881 hydrochloride: Potent, selective inhibitor of VEGF-2
## Regulators

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<th>Name</th>
<th>Description</th>
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<td>Difopein</td>
<td>High affinity inhibitor of 14.3.3 proteins; induces apoptosis</td>
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<tr>
<td>2144</td>
<td>R18</td>
<td>Inhibitor of 14.3.3 proteins; induces apoptosis</td>
</tr>
<tr>
<td>Src 1629</td>
<td>Herbimycin A</td>
<td>Src family kinase inhibitor. Also Hsp90 inhibitor</td>
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<td>1397</td>
<td>PP-1</td>
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<td>3063</td>
<td>1-Naphthyl PP1</td>
<td>Src family kinase inhibitor; also inhibits c-Abl</td>
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<td>Hsp90 1515</td>
<td>17-AAG</td>
<td>Selective Hsp90 inhibitor</td>
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<tr>
<td>2435</td>
<td>CCT 018159</td>
<td>Hsp90 inhibitor</td>
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<tr>
<td>1368</td>
<td>Geldanamycin</td>
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<tr>
<td>1629</td>
<td>Herbimycin A</td>
<td>Hsp90 inhibitor. Also Src family kinase inhibitor</td>
</tr>
<tr>
<td>1589</td>
<td>Radicicol</td>
<td>Hsp90 inhibitor. Antifungal antibiotic</td>
</tr>
<tr>
<td>SGK 3572</td>
<td>GSK 650394</td>
<td>Serine- and glucocorticoid-regulated kinase (SGK) inhibitor</td>
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<tr>
<td>3622</td>
<td>IPA 3</td>
<td>Group I p21-activated kinase (PAK) inhibitor</td>
</tr>
<tr>
<td>PAK1 1336</td>
<td>Calyculin A</td>
<td>Protein phosphatase 1 and 2A inhibitor</td>
</tr>
<tr>
<td>1840</td>
<td>Fostriecin sodium salt</td>
<td>Potent PP2A and PP4 inhibitor</td>
</tr>
<tr>
<td>1136</td>
<td>Okadaic acid</td>
<td>Protein phosphatase 1 and 2A inhibitor</td>
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</tbody>
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## Downstream targets and effectors

<table>
<thead>
<tr>
<th>Code</th>
<th>Name</th>
<th>Description</th>
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</thead>
<tbody>
<tr>
<td>1290</td>
<td>Anisomycin</td>
<td>Activates JNK/SAPK/p38 MAP kinase</td>
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<tr>
<td>2731</td>
<td>CGP 57380</td>
<td>Selective inhibitor of Mnk1</td>
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<tr>
<td>1989</td>
<td>c-JUN peptide</td>
<td>JNK/c-Jun interaction inhibitor</td>
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<td>2358</td>
<td>Anti-c-Jun (Clone CJ 4C4/1)</td>
<td>Antibody recognizing c-Jun</td>
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<tr>
<td>3140</td>
<td>PHA 767491 hydrochloride</td>
<td>MK-2 inhibitor. Also inhibits cdc7/cdk9</td>
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<td>2250</td>
<td>SL 0101-1</td>
<td>Selective p90 ribosomal S6 kinase (RSK) inhibitor</td>
</tr>
<tr>
<td>2476</td>
<td>SR 11302</td>
<td>Inhibitor of AP-1 transcription factor; antitumor agent</td>
</tr>
</tbody>
</table>

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