

RESEARCH MONOGRAPH . COMPOUND PROFILE

Fasudil

An isoquinolinesulfonamide kinase inhibitor developed by Asahi Kasei as the first clinically approved ROCK inhibitor, registered in Japan and China for cerebral vasospasm after subarachnoid hemorrhage, with expanding investigational applications in pulmonary hypertension, amyotrophic lateral sclerosis, and cardiovascular disease.

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ABSTRACT

Fasudil (HA-1077, AT-877), the isoquinolinesulfonamide derivative of the H-series protein kinase inhibitors first described by Hidaka et al. in 1984, is the only Rho-associated coiled-coil-containing protein kinase (ROCK) inhibitor approved for systemic clinical use and the first protein kinase inhibitor to receive regulatory approval for any indication. Developed by Asahi Kasei Pharma (Tokyo, Japan) and marketed as Eril, fasudil has been approved in Japan since 1995 and in China for the treatment of cerebral vasospasm following aneurysmal subarachnoid hemorrhage, where it is standard of care in Japanese neurosurgical practice. The compound inhibits ROCK by competitive occupation of the ATP-binding domain, with a K_i of approximately 0.33 micromolar against ROCK activity and modest preferential selectivity for ROCK2 (K_i approximately 47 nanomolar) over ROCK1 (K_i approximately 76 nanomolar) in purified kinase assays. Off-target inhibition of protein kinase A, protein kinase G, protein kinase C, and myosin light chain kinase occurs at approximately 5- to 100-fold higher concentrations. The principal metabolic transformation is rapid hepatic conversion to hydroxyfasudil by aldehyde oxidase (not cytochrome P450), producing an active metabolite with ROCK inhibitory potency similar to or greater than the parent compound and a substantially longer plasma half-life (approximately 5.5 hours versus 0.55 hours for fasudil). This metabolic profile renders fasudil effectively a prodrug of hydroxyfasudil in the oral context. The clinical evidence base in cerebral vasospasm rests on a pivotal 267-patient double-blind placebo-controlled trial (Shibuya et al. 1992) demonstrating significant reductions in angiographic vasospasm (38 versus 61

percent), symptomatic vasospasm (35 versus 50 percent), and poor outcome on the Glasgow Outcome Scale (12 versus 26 percent), confirmed by a 1,462-patient postmarketing surveillance study and a meta-analysis of 8 trials (odds ratio 0.48 for symptomatic vasospasm). Head-to-head comparison with nimodipine demonstrated comparable or superior outcomes (74.5 versus 61.7 percent good clinical outcome). Expanding investigational programs span pulmonary arterial hypertension, where meta-analyses of up to 865 patients demonstrate significant reductions in pulmonary artery pressure, vascular resistance, and 6-minute walk distance with no serious adverse events; and amyotrophic lateral sclerosis, where the 120-patient ROCK-ALS Phase 2 trial confirmed safety, demonstrated significant electrophysiological biomarker preservation (motor unit number index), and confirmed cerebrospinal fluid penetration of the active metabolite at concentrations matching preclinical efficacy. This monograph reviews the chemistry, synthesis, and structure-activity relationships of fasudil; the kinase inhibition profile and downstream signaling including ROCK-mediated myosin light chain phosphorylation, NF-kappaB inflammatory signaling, PPARalpha-NOX neuroprotective axis, and microglial modulation; the comprehensive pharmacokinetics of fasudil and hydroxyfasudil from the first formal Phase I oral bioavailability study; the preclinical evidence across cerebrovascular, cardiovascular, spinal cord injury, and neurodegenerative disease models; the full clinical evidence base across cerebral vasospasm, pulmonary hypertension, amyotrophic lateral sclerosis, and coronary vasospasm indications; and a comparative assessment against four ROCK inhibitor comparators (Y-27632, ripasudil, netarsudil, belumosudil) and the calcium channel blocker nimodipine. Fasudil is approved only in Japan and China. It is

available as a research-grade preparation from established chemical suppliers; investigators should obtain analytical confirmation of identity and purity on every lot.

1. Compound Identification

1.1 Names, synonyms, and registry identifiers

International Nonproprietary Name: Fasudil.

Marketed name: Eril (Asahi Kasei Pharma; Japan).

Development codes: HA-1077 (Hidaka series designation, identifying the compound within the H-series of isoquinolinesulfonamide protein kinase inhibitors), AT-877 (Asahi Kasei clinical development code, appearing in the pivotal Japanese clinical trial publications), ZK-258594 [1, 2, 4]. The HA-1077 designation is the most common identifier in the pharmacological literature; AT-877 appears principally in the cerebral vasospasm clinical trial reports.

Chemical Abstracts Service registry number: 103745-39-7 (free base). The hydrochloride salt and its hydrate (fasudil hydrochloride hydrate) are the clinically formulated forms; the hydrate is the form used in the marketed intravenous preparation (Eril).

International Union of Pure and Applied Chemistry name: 5-(1,4-diazepan-1-ylsulfonyl)isoquinoline [PubChem CID 3547]. The compound is also referred to in the pharmacological literature as 1-(5-isoquinolinesulfonyl)homopiperazine, reflecting the two-ring structural description: an isoquinoline aromatic system connected through a sulfonamide linkage to a homopiperazine (1,4-diazepane) ring [2].

Database identifiers: PubChem CID 3547; ChEMBL CHEMBL38380; DrugBank DB08162; UNII Q0CH43PGXS; ATC code C04AX32 (other peripheral vasodilators).

1.2 Structural class and chemistry

Fasudil is an isoquinolinesulfonamide, a structural class first described by the Japanese biochemist Hiroyoshi Hidaka and colleagues at Mie University in 1984 as synthetic, cell-permeable protein kinase inhibitors with sufficient affinity to compete against endogenous ATP at the kinase catalytic domain [1, 43]. The compound consists of an isoquinoline aromatic ring system connected through a sulfonamide bond to a seven-membered homopiperazine ring. This architecture places fasudil

within the H-series of isoquinolinesulfonamides, which includes H-7, H-8, H-9, and H-89, all sharing the isoquinoline sulfonamide core with different cyclic amine substituents [2].

Molecular formula: C₁₄H₁₇N₃O₂S (free base).

Exact mass: 291.10415 g/mol (monoisotopic) [PubChem CID 3547].

Molecular weight: 291.37 g/mol (free base) [PubChem CID 3547].

Canonical SMILES: C1CNCCN(C1)S(=O)(=O)C2=CC=CC3=C2C=CN=C3

InChIKey: NGOGFTYYXHNFQH-UHFFFAOYSA-N

Physical properties: White crystalline powder. The hydrochloride salt is freely soluble in water. The XLogP of the free base is 1, indicating moderate hydrophilicity [PubChem CID 3547]. The compound lacks chiral centers and is supplied as a single achiral chemical entity; no enantiomeric or diastereomeric considerations apply.

1.3 Synthesis

The classical synthesis of fasudil proceeds from 5-isoquinolinesulfonyl chloride and homopiperazine (1,4-diazepane) via direct sulfonamidation in the presence of a base [33]. An alternative six-step process starting from ethylenediamine and 5-isoquinolinesulfonyl chloride avoids the use of costly pre-formed homopiperazine through sequential sulfonamidation, protection, nucleophilic substitution, deprotection, cyclization, and salification [44]. The optimized one-step sulfonamidation approach achieves yields of approximately 73 percent [34].

Structure-activity relationship studies by Loge et al. (2003) demonstrated that both the isoquinolinyl nitrogen and the basic amino group separated by a sulfonamide-bearing spacer are essential for ROCK inhibitory activity; most modifications to either the heterocycle or the sulfonyl group resulted in activity loss [33]. The single analog that matched fasudil's potency while improving selectivity was a tetrahydroisoquinoline derivative, which lost the ability to inhibit protein kinase C, suggesting that the fully aromatic isoquinoline ring is responsible for the PKC off-target activity [33]. The hydrochloride salt is prepared by treatment of the free base with hydrochloric acid; the hydrate form (fasudil hydrochloride hydrate) is obtained by controlled crystallization and is the marketed pharmaceutical form.

2. Abstract

The abstract appears in the YAML frontmatter above and is not repeated as a body section.

3. Discovery and Development History

3.1 Origin

Fasudil was developed as part of the H-series of isoquinolinesulfonamide protein kinase inhibitors by Hiroyoshi Hidaka at Mie University School of Medicine. In October 1984, Hidaka and colleagues published the first report of isoquinolinesulfonamide compounds as potent, cell-permeable inhibitors of cyclic nucleotide-dependent protein kinases (PKA, PKG) and protein kinase C, establishing that the ATP-binding site of kinase catalytic domains was druggable by small molecules [1, 43]. Among the series, the compound designated HA-1077 (1-(5-isoquinolinesulfonyl)homopiperazine) was identified by Asahi Chemical Industry (now Asahi Kasei Pharma, Tokyo, Japan) as a vasodilator that preferentially suppressed cerebral vasospasm without significant changes in systemic blood pressure [2]. Fasudil was first published in 1986 as a calcium antagonist that attenuated experimentally induced cerebral vasospasm in dogs. Asahi Chemical Industry undertook clinical development under the code AT-877.

3.2 Identification as ROCK inhibitor

The initial pharmacological characterization classified fasudil as an intracellular calcium antagonist and protein kinase inhibitor. The specific identification of Rho-associated protein kinase (ROCK) as the principal pharmacological target at clinically relevant concentrations emerged from the work of Uehata et al. (1997), published in *Nature* as "Calcium sensitization of smooth muscle mediated by a Rho-associated protein kinase in hypertension" [3]. That study identified the Rho-associated kinase p160ROCK as the target mediating calcium sensitization of vascular smooth muscle and demonstrated that pharmacological inhibition of this kinase normalized blood pressure in hypertensive rat models. Subsequent characterization of fasudil's kinase inhibition profile confirmed that ROCK was the most potently inhibited kinase at therapeutic concentrations, with a K_i at least 10-fold lower than that for other AGC-family kinases [2]. This reclassification from "calcium antagonist" to "ROCK inhibitor" transformed the pharmacological understanding of fasudil's therapeutic mechanism and catalyzed a broad expansion of the research literature.

3.3 Clinical approval and registration

In September 1995, fasudil hydrochloride hydrate (Eril) received marketing approval in Japan for the prevention and treatment of cerebral vasospasm following aneurysmal subarachnoid hemorrhage [4]. The approval was supported by the pivotal placebo-controlled double-blind trial conducted by Shibuya et al. across 60 Japanese neurosurgical centers [4]. The compound was subsequently approved in China for the same indication. Fasudil is not approved by the United States Food and Drug Administration, the European Medicines Agency, or other Western regulatory authorities. No oral formulation is currently approved for any indication, though Phase I pharmacokinetic data for oral fasudil have been published [15]. Fasudil holds the distinction of being the first protein kinase inhibitor approved for clinical use; more than 70 kinase inhibitors have subsequently received regulatory approval worldwide [43].

3.4 Expansion of the research literature

The identification of ROCK as a central regulator of vascular tone, cell migration, inflammation, and neuronal survival has driven investigational interest in fasudil across multiple therapeutic areas. Published clinical or preclinical investigations now span pulmonary arterial hypertension [9, 10], coronary vasospasm and stable angina [31, 38], amyotrophic lateral sclerosis [13, 14, 21], Alzheimer's disease [24], spinal cord injury [22], ischemic stroke [19, 20], myocardial ischemia-reperfusion injury [25], and stem cell biology [34]. A 2023 Japanese nationwide survey of 158 neurosurgical hospitals confirmed that 100 percent of responding institutions use fasudil for vasospasm prophylaxis after aneurysmal subarachnoid hemorrhage, confirming its status as standard of care in Japan [29].

4. Mechanism of Action

Fasudil is a competitive inhibitor of Rho-associated coiled-coil-containing protein kinase (ROCK), binding at the ATP-binding pocket of the kinase catalytic domain. ROCK exists as two isoforms, ROCK1 and ROCK2, that serve as principal downstream effectors of the small GTPase RhoA. The RhoA/ROCK signaling axis is a central regulator of smooth muscle contraction, cytoskeletal dynamics, cell migration, inflammatory signaling, and neuronal survival. Fasudil inhibits both isoforms, with modest preferential activity at ROCK2.

4.1 ROCK inhibition: binding parameters and isoform selectivity

Published K_i and IC_{50} values for fasudil vary by assay system and source. In cell-free kinase assays, fasudil displays a K_i of approximately 0.33 micromolar against ROCK activity [2, 40]. More recent measurements using purified recombinant kinase domains report K_i values of 76 nanomolar for ROCK1 and 47 nanomolar for ROCK2, confirming modest ROCK2 preference [35]. In cell-based assays, IC_{50} values are higher: approximately 1.9 to 3.7 micromolar for ROCK, reflecting the contribution of intracellular ATP concentration (approximately 1 to 5 millimolar) to competitive displacement at the binding site [35]. The difference between cell-free K_i and cell-based IC_{50} is expected for an ATP-competitive inhibitor and should be considered when extrapolating from biochemical potency to cellular efficacy.

4.2 Selectivity profile

Fasudil was originally developed as an inhibitor of cyclic nucleotide-dependent protein kinases and protein kinase C, and retains measurable activity against these targets at higher concentrations [1, 2]. Published K_i values for off-target kinases in cell-free assays are: protein kinase A (PKA) 1.6 micromolar, protein kinase G (PKG) 1.6 micromolar, protein kinase C (PKC) 3.3 micromolar, and myosin light chain kinase (MLCK) 36 micromolar [2, 40]. Additional off-target activity has been reported against PKN2 (IC_{50} approximately 4 micromolar) and MAPK1 (IC_{50} approximately 5 micromolar). Fasudil was described as non-selective against 8 of 27 kinases tested in one profiling panel, a broader off-target footprint than the research tool com-

pound Y-27632 (non-selective against 4 of 25 tested kinases) [35]. Despite this, the at least 5- to 10-fold potency differential between ROCK and the nearest off-target kinases (PKA, PKG) ensures that ROCK is the dominant target at clinically achieved plasma concentrations.

4.3 Active metabolite: hydroxyfasudil

Fasudil is rapidly converted in vivo to hydroxyfasudil, which retains ROCK inhibitory activity with similar or greater potency than the parent compound and accounts for the majority of the sustained pharmacological effect [2, 15, 16]. Hydroxyfasudil has been reported as more selective for ROCK over PKA than fasudil, contributing to a cleaner pharmacological profile at steady state [35]. The half-life of hydroxyfasudil in humans is approximately 5.5 to 5.7 hours, substantially longer than the parent compound (approximately 0.55 hours), meaning that the active metabolite is the dominant circulating species for the majority of the dosing interval [15].

4.4 Downstream signaling

ROCK phosphorylates a cascade of substrates that regulate smooth muscle tone, cytoskeletal organization, and inflammatory signaling.

Myosin light chain and MLC phosphatase: ROCK directly phosphorylates myosin light chain (MLC) at Ser19 and also phosphorylates MYPT1 (the regulatory subunit of MLC phosphatase), inactivating the phosphatase. The net effect is sustained MLC phosphorylation and smooth muscle contraction independent of intracellular calcium concentration [25, 26]. Fasudil, by inhibiting ROCK, reduces both direct MLC phosphorylation and MYPT1 phosphorylation, promoting vascular smooth muscle relaxation. This calcium sensitization mechanism is the molecular basis for the vasodilatory effects in cerebral vasospasm, coronary vasospasm, and pulmonary hypertension.

LIM kinase and cofilin: ROCK phosphorylates LIM kinase, which in turn phosphorylates cofilin, stabilizing actin filaments and promoting stress fiber formation. Fasudil-mediated ROCK inhibition destabilizes actin stress fibers and promotes cytoskeletal remodeling, with consequences for cell migration, neurite outgrowth, and axonal regeneration [22, 40].

eNOS and PI3K/Akt: ROCK negatively regulates endothelial nitric oxide synthase (eNOS) through multiple mechanisms. Fasudil increases eNOS activity and nitric oxide bioavailability, contributing to vasodilation and endothelial protection [25, 32].

NF-kappaB signaling: The RhoA/ROCK pathway activates NF-kappaB-dependent transcription of proinflammatory cytokines. In lipopolysaccharide-stimulated rat pulmonary microvascular endothelial cells, fasudil suppressed NF-kappaB p65 nuclear translocation, reducing expression of interleukin-6 and monocyte chemoattractant protein-1 and lowering reactive oxygen species levels [27].

ERK/YAP signaling: In hepatic artery smooth muscle, fasudil inhibits the YAP/ERK/ETA/ETB signaling cascade. ROCK serves as an upstream activator of ERK1/2, which in turn activates the Hippo/YAP pathway and endothelin receptor signaling, a cascade implicated in vasospasm. Fasudil at 1.4 mg/kg daily for 7 days in rabbits and at 30 micromolar in vitro significantly reduced RhoA, ROCK, and phospho-ERK1/2 expression [26].

4.5 Neuroprotective mechanisms

The neuroprotective activity of fasudil operates through several ROCK-dependent pathways beyond vasodilation.

ROCK-PPARalpha-NOX axis: In a mouse middle cerebral artery occlusion model, fasudil at 30 mg/kg by gavage reduced cerebral infarction volume by approximately 48 percent (52.2 plus or minus 7.6 versus 99.9 plus or minus 4.8 cubic millimeters; P less than 0.01). The mechanism involved upregulation of PPARalpha (from 0.614 to 1.417 relative expression), downregulation of NOX2 mRNA and NADPH oxidase activity (to approximately 49 percent of control), and reduction of reactive oxygen species content (to approximately 47 percent of control). The neuroprotective effect was abolished in PPARalpha knockout mice, confirming pathway dependence [20].

Microglial modulation: ROCK II expression and activity in brain tissue are selectively upregulated in microglia (not in neurons or astrocytes) after hypoxia-reoxygenation injury. Fasudil inhibits microglial ROCK activity, reduces inducible nitric oxide synthase, and shifts the cytokine profile from proinflammatory (reduced nitric oxide, interleukin-1beta, interleukin-6, tumor necrosis factor alpha) to anti-in-

flammatory (increased interleukin-10) [23]. This microglial-specific mechanism is the molecular basis for the neuroprotection observed in ischemia and neurodegeneration models.

4.6 Composite pharmacology at clinical concentrations

At the registered intravenous dose of 30 mg three times daily for cerebral vasospasm, peak serum concentrations of fasudil reach approximately 100 to 280 nanograms per milliliter (0.34 to 0.96 micromolar), declining rapidly with a half-life of approximately 0.55 hours [9, 15]. The dominant circulating species within 1 to 2 hours becomes hydroxyfasudil, with peak concentrations of approximately 108 to 173 nanograms per milliliter and a half-life of approximately 5.5 hours [9, 15]. At these concentrations, substantial ROCK inhibition is achieved, with partial inhibition of PKA and PKG and minimal inhibition of PKC and MLCK. The selectivity for pulmonary over systemic vasculature is a clinically important observation: intravenous fasudil at 30 mg reduced pulmonary vascular resistance by 17 percent in patients with severe pulmonary hypertension without producing systemic hypotension [9]. This selectivity likely reflects elevated baseline ROCK activity in the pulmonary vasculature under pathological conditions, where ROCK-mediated calcium sensitization is disproportionately driving vascular tone.

5. Pharmacokinetics and Routes of Administration

5.1 Absorption

Fasudil hydrochloride is rapidly absorbed after oral administration but undergoes near-complete first-pass hepatic metabolism. In the SAFE-ROCK Phase I trial in 14 healthy volunteers, a single 30 mg oral dose produced a median Tmax of 0.25 hours (range 0.00 to 0.50 hours) for the parent compound, with peak plasma concentrations (Cmax) of only 1.4 micrograms per liter (coefficient of variation 41.0 percent); 28.6 percent of subjects had no detectable fasudil in plasma after oral dosing [15]. In contrast, the active metabolite hydroxyfasudil reached a Cmax of 111.6 micrograms per liter (CV 24.1 percent) at a median Tmax of 0.50 hours [15]. By comparison, a 30 mg intravenous infusion over 30 minutes produced a fasudil Cmax of 100.6 micrograms per liter (CV 74.2 percent) and a hydroxyfasudil Cmax of 108.4 micrograms per liter (CV 19.7 percent) [15].

The absolute bioavailability of hydroxyfasudil after oral fasudil was approximately 69 percent relative to intravenous administration, calculated from AUC comparisons (oral AUC0-infinity 310.3 versus intravenous 459.8 micrograms times hours per liter) [15]. A crossover study in 8 healthy volunteers administering fasudil to different gastrointestinal sites (oral solution, oral tablet, distal ileum, ascending colon as solution, ascending colon as powder) found similar hydroxyfasudil systemic availability across all sites (relative bioavailability 1.04 to 1.27), supporting the feasibility of extended-release oral formulation development [17].

5.2 Metabolism

Fasudil undergoes rapid and extensive hepatic metabolism. The principal metabolic transformation is hydroxylation to hydroxyfasudil, catalyzed by aldehyde oxidase (AO) rather than cytochrome P450 enzymes or xanthine oxidase [16]. The oxygen atom incorporated into hydroxyfasudil originates from water rather than atmospheric oxygen, confirming the aldehyde oxidase mechanism [16]. Fasudil has a higher affinity for human hepatic AO than for rat hepatic AO; hydroxyfasudil is generated in large amounts by both rat and human liver cytosol but is not detected in

dog liver cytosol, indicating a species-specific metabolic pathway relevant to pre-clinical-to-clinical translation [16]. Gender-dependent aldehyde oxidase activity has been observed in rats, producing significantly different pharmacokinetic parameters between male and female animals; the clinical relevance to human sex differences has not been formally established [16, 18]. The rapid first-pass conversion explains the near-absence of parent compound in plasma after oral dosing and has led investigators to characterize fasudil as effectively a prodrug of hydroxyfasudil in the oral context [15].

5.3 Distribution

Fasudil distributes widely into tissues. In preclinical studies in rats and dogs, AUC and C_{max} showed dose-proportional increases within the tested range of 2 to 6 mg/kg [18]. Central nervous system penetration is a critical property. In the ROCK-ALS Phase 2 trial, cerebrospinal fluid concentrations of hydroxyfasudil were 5.8 plus or minus 2.0 micrograms per liter (30 mg group) and 9.4 plus or minus 4.5 micrograms per liter (60 mg group), values similar to concentrations achieved in the SOD1G93A mouse model at pharmacologically effective doses (8.8 plus or minus 3.0 micrograms per liter) [13]. In the PS19 tau transgenic mouse model, significant brain concentrations of both fasudil and hydroxyfasudil were detected after intraperitoneal administration, with a negative correlation between brain drug levels and phosphorylated tau at residue 396 [24]. These data confirm that the compound and its active metabolite reach the CNS at concentrations exceeding the K_i for ROCK, supporting the pharmacological basis for neuroprotective effects.

5.4 Elimination

Fasudil has a very short plasma elimination half-life of approximately 0.55 hours after intravenous administration in healthy volunteers [15]. Hydroxyfasudil has a considerably longer half-life of 5.54 hours (oral) to 5.70 hours (intravenous), with no significant route-dependent difference (P equals 0.50) [15]. Elimination is predominantly metabolic (hepatic aldehyde oxidase-mediated biotransformation) rather than direct renal clearance of the parent compound. The high inter-individual variability in fasudil C_{max} (coefficient of variation 55 to 74 percent across studies) is attributable to the rapid metabolism and individual differences in aldehyde oxidase activity [15].

5.5 Routes of administration

The registered route is intravenous infusion: 30 mg fasudil hydrochloride hydrate in solution, administered over 30 minutes, three times daily for 14 days (cerebral vasospasm indication) [4]. Intracoronary administration (for coronary vasospasm research) and intra-arterial infusion (for post-traumatic cerebral vasospasm) have been reported in clinical studies. Oral administration has been evaluated in the SAFE-ROCK Phase I trial at doses of 30 mg single and 90 mg daily (30 mg three times daily), with good tolerability and no serious adverse events [15].

5.6 Special populations

Gender differences in fasudil pharmacokinetics have been documented in rats (absolute bioavailability and excretion) but not in dogs [18]. Hepatic impairment would be expected to alter fasudil metabolism given the dependence on hepatic aldehyde oxidase; no formal hepatic impairment study has been published. Pediatric and geriatric pharmacokinetics have not been formally characterized in published studies.

5.7 Pharmacokinetic parameters from clinical studies

In patients with severe pulmonary hypertension receiving 30 mg intravenous fasudil over 30 minutes, Fukumoto et al. (2005) reported peak serum concentrations of 278.9 (standard deviation 150.5) nanograms per milliliter of fasudil and 173.2 (10.3) nanograms per milliliter of hydroxyfasudil [9]. The serum half-life of fasudil after intravenous injection has been independently reported as approximately 20 minutes (0.33 hours) in neurosurgical patients, consistent with the 0.55 hours measured in the SAFE-ROCK healthy-volunteer study [15]. Oral dosing at 90 mg daily (30 mg three times daily) in the SAFE-ROCK trial was well tolerated; hydroxyfasudil achieved sustained concentrations across the dosing interval [15].

6. Preclinical Pharmacology

6.1 Cerebrovascular and ischemic neuroprotection

The neuroprotective activity of fasudil was demonstrated by Satoh et al. (1996) in two species and two models [19]. In a gerbil model of transient bilateral carotid artery occlusion, fasudil at 30 mg/kg preserved hippocampal neuronal density at 101.0 plus or minus 22.0 cells per millimeter, compared to 17.8 plus or minus 2.1 cells per millimeter in controls, a nearly six-fold difference. Neither nimodipine nor ozagrel sodium produced significant neuronal protection in the same model [19]. In a rat model of cerebral microembolization by microsphere injection, fasudil dose-dependently reduced cortical infarct area: 21.5 plus or minus 4.8 percent of cortical section at 10 mg/kg versus 43.9 plus or minus 2.4 percent in controls, with intermediate reduction at 3 mg/kg (32.7 plus or minus 2.8 percent). Brain edema was similarly reduced: 81.0 plus or minus 0.4 percent water content versus 82.4 plus or minus 0.2 percent in controls [19].

In a mouse middle cerebral artery occlusion (MCAO) model, fasudil at 30 mg/kg by oral gavage for three days prior to ischemia-reperfusion reduced cerebral infarct volume by approximately 48 percent (52.2 plus or minus 7.6 versus 99.9 plus or minus 4.8 cubic millimeters; P less than 0.01), operating through the ROCK-PPARalpha-NOX axis in a PPARalpha-dependent manner [20].

6.2 Cardioprotection

A meta-analysis by Huang et al. (2018) pooled 19 preclinical studies involving 400 animals evaluating fasudil in myocardial ischemia-reperfusion injury models across rats, mice, rabbits, and swine [25]. Fasudil treatment significantly reduced myocardial infarct area relative to left ventricular area (standardized mean difference negative 3.25, 95 percent confidence interval negative 3.75 to negative 2.74; P less than 0.00001) and infarct area relative to area at risk (standardized mean difference negative 2.60, 95 percent CI negative 3.35 to negative 1.85; P less than 0.00001) [25]. Serum creatine kinase was also significantly reduced (standardized mean difference negative 4.43, P less than 0.00001, I-squared 0 percent). Systolic and diastolic cardiac function were improved (P less than 0.05). Nine mechanistic pathways

were identified in the meta-analysis: coronary vasodilation via PI3K/Akt/eNOS up-regulation, anti-apoptosis through Bcl-2 upregulation and caspase-3 downregulation, anti-inflammatory cytokine suppression, antioxidant enhancement (SOD activity), endoplasmic reticulum stress reduction, ATP-competitive ROCK inhibition, mitochondrial K-ATP channel activation, and autophagy improvement [25]. Both pre-ischemia and early reperfusion administration were effective, with no significant difference in effect size by timing of administration.

6.3 Spinal cord injury

Impellizzeri et al. (2012) evaluated fasudil at 10 mg/kg intraperitoneal (at 1 and 6 hours post-injury) in a mouse spinal cord compression model [22]. Fasudil significantly reduced histological damage, motor impairment, NF-kappaB expression, inflammasome activation (caspase-1, NLRP3), proinflammatory cytokines (tumor necrosis factor alpha, interleukin-1beta), neutrophil infiltration, nitrosative stress markers, glial activation, apoptosis, and MAPK pathway activation (phospho-ERK, phospho-JNK). The authors concluded that ROCK inhibition by fasudil "may represent a useful therapeutic perspective in the treatment of inflammation associated with spinal cord trauma" [22].

6.4 Amyotrophic lateral sclerosis models

Gunther et al. (2017) treated SOD1G93A transgenic mice with oral fasudil at 30 and 100 mg/kg/day in drinking water beginning at symptomatic stage (day 80 of life) [21]. Fasudil significantly improved motor behavior in male mice, though survival was not significantly extended when treatment began after symptom onset. The authors noted that "treatment after symptom onset was less potent than treatment in asymptomatic animals," indicating that the timing of ROCK inhibition relative to disease stage affects efficacy [21]. Earlier work demonstrated that presymptomatic fasudil treatment limited motor neuron loss and modulated microglial activation in ALS mouse models [41, 42].

6.5 Alzheimer's disease models

Collu et al. (2024) treated PS19 tau transgenic mice (expressing P301S mutant human MAPT) with fasudil at 30 and 100 mg/kg/day by intraperitoneal injection for two weeks beginning at 6 months of age [24]. Brain concentrations of fasudil and hydroxyfasudil showed a significant negative correlation with phosphorylated tau

at residue 396. Proteomic analysis revealed activation of mitochondrial tricarboxylic acid cycle and blood-brain barrier gap junction metabolic pathways [24]. These findings demonstrate that even short-duration fasudil treatment engages tau phosphorylation pathways and produces measurable brain proteomic changes in a model relevant to Alzheimer's disease.

6.6 Hippocampal neuroprotection

Ding et al. (2010) demonstrated that fasudil protects hippocampal neurons against hypoxia-reoxygenation injury in mice through suppression of microglial inflammatory responses [23]. ROCK II expression and activity were specifically upregulated in microglia (not in neurons or astrocytes) after hypoxia-reoxygenation. Fasudil inhibited microglial ROCK activity, reduced inducible nitric oxide synthase, and shifted the cytokine profile toward anti-inflammatory: reduced nitric oxide, interleukin-1beta, interleukin-6, and tumor necrosis factor alpha, with increased interleukin-10 [23]. This finding established that fasudil's neuroprotection operates primarily through microglial modulation rather than direct neuronal effects.

6.7 Anti-inflammatory pharmacology

In rat pulmonary microvascular endothelial cells stimulated with lipopolysaccharide, fasudil at 10 to 50 micromolar suppressed NF-kappaB p65 nuclear translocation, reduced reactive oxygen species levels, and reduced interleukin-6 and monocyte chemoattractant protein-1 mRNA expression [27]. In a rabbit hepatic artery spasm model, fasudil at 1.4 mg/kg daily for 7 days repressed the YAP/ERK/ETA/ETB signaling pathway, with significant reductions in RhoA, ROCK, and phospho-ERK1/2 expression in arterial smooth muscle [26].

6.8 Stem cell biology

Fasudil has been validated as a functionally equivalent, cost-effective replacement for Y-27632 in human pluripotent stem cell research. So et al. (2020) demonstrated that fasudil at 10 micromolar promoted comparable survival, anti-apoptotic protection, and differentiation capacity to Y-27632 in human embryonic and induced pluripotent stem cells [34]. Cell survival after freeze-thaw over 7 days was 8.1-fold (fasudil) versus 6.7-fold (Y-27632) in hES2 cells (P less than 0.05); apoptosis rates

were equivalent at 34 percent for both inhibitors at 24 hours [34]. Fasudil is 10 to 30 times cheaper than Y-27632, reflecting its simpler one-step synthesis (73 percent yield) versus the seven-step synthesis required for Y-27632 (45 percent yield) [34].

7. Clinical Evidence Base

The following clinical evidence is presented for research context. Fasudil is approved only in Japan and China for cerebral vasospasm following subarachnoid hemorrhage; all other clinical applications described below are investigational.

7.1 Cerebral vasospasm after subarachnoid hemorrhage

The pivotal trial was a prospective, randomized, placebo-controlled, double-blind multicenter study across 60 Japanese neurosurgical centers (Shibuya et al. 1992) [4]. A total of 267 patients with Hunt and Hess grades I through IV subarachnoid hemorrhage who had undergone surgical clipping within 3 days were randomized to fasudil (AT-877) 30 mg intravenously over 30 minutes three times daily for 14 days or placebo. Fasudil reduced angiographic vasospasm from 61 percent to 38 percent (P equals 0.0023), CT low-density regions from 38 percent to 16 percent (P equals 0.0013), symptomatic vasospasm from 50 percent to 35 percent (P equals 0.0247), and poor outcome on the Glasgow Outcome Scale from 26 percent to 12 percent (P equals 0.0152). No serious adverse events were reported [4].

Zhao et al. (2006) randomized 72 patients to fasudil 30 mg intravenous three times daily for 14 days versus nimodipine 1 mg/hour continuous infusion [5]. Symptomatic vasospasm occurred in 5 of 33 fasudil patients versus 9 of 32 nimodipine patients; good recovery on the Glasgow Outcome Scale was achieved in 23 of 33 fasudil patients versus 19 of 34 nimodipine patients. No serious adverse events were reported in the fasudil group [5]. The final results of the Fasudil Aneurysmal Subarachnoid Hemorrhage Study Group (Zhao et al. 2011) extended this comparison to 129 patients (63 fasudil, 66 nimodipine) and reported good clinical outcome in 74.5 percent (41 of 55) of fasudil patients versus 61.7 percent (37 of 60) of nimodipine patients (P equals 0.040) [6].

A postmarketing surveillance study by Suzuki et al. (2007) evaluated 1,462 patients treated with fasudil between 1995 and 2000, confirming the safety profile of the pivotal trial. The absence of symptomatic vasospasm was more common in the postmarketing population than in the phase 3 trial [7].

A meta-analysis by Liu et al. (2012) pooling 8 randomized controlled trials found that fasudil approximately halved the odds of symptomatic cerebral vasospasm (odds ratio 0.48, 95 percent CI 0.32 to 0.72; P equals 0.0005), angiographic vasospasm (odds ratio 0.40, 95 percent CI 0.24 to 0.67; P equals 0.0004), and cerebral infarction (odds ratio 0.50, 95 percent CI 0.34 to 0.76; P equals 0.0009) [8].

A 2023 Japanese nationwide survey of 158 hospitals (3,093 patients) confirmed that all responding institutions use fasudil for vasospasm prophylaxis. The most effective combination was cilostazol, fasudil, and statin (odds ratio 0.38, 95 percent CI 0.22 to 0.67; P equals 0.005), with an overall delayed cerebral infarction incidence of 9.1 percent [29]. In the J-ASPECT database study of 17,343 patients across 579 hospitals, fasudil use in elderly patients (aged 75 and older) was associated with reduced in-hospital mortality (odds ratio 0.33, 95 percent CI 0.25 to 0.42; P less than 0.01), though improvement in functional outcomes did not reach significance (odds ratio 0.85, 95 percent CI 0.66 to 1.08; P equals 0.19) [30].

7.2 Pulmonary hypertension

Fukumoto et al. (2005) reported the acute hemodynamic effects of intravenous fasudil 30 mg over 30 minutes in 9 patients with severe pulmonary hypertension (baseline mean pulmonary artery pressure 58 mmHg, baseline pulmonary vascular resistance index 2429 dyne times seconds times centimeters to the negative fifth times meters squared). Fasudil significantly reduced pulmonary vascular resistance by 17 percent without systemic hypotension [9].

A systematic review by Abedi et al. (2023) including 12 studies (575 patients, 8 short-term and 4 mid-term trials) found significant improvement in mean pulmonary artery pressure, systolic pulmonary artery pressure, pulmonary vascular resistance, systemic vascular resistance, and cardiac index across short-term trials. One mid-term trial demonstrated that fasudil significantly decreased rehospitalization and mortality. No serious adverse effects were reported [10].

A 2026 meta-analysis of 11 randomized controlled trials (865 participants) with COPD-associated pulmonary arterial hypertension found fasudil improved the overall effective rate (risk ratio 1.18, 95 percent CI 1.05 to 1.31; P equals 0.004), reduced pulmonary artery systolic pressure by 9.42 mmHg (95 percent CI negative 10.73 to negative 8.12; P less than 0.001), and increased 6-minute walk distance by 51.96 meters (95 percent CI 36.84 to 67.08; P less than 0.001) [11]. Chronic treatment

lasting two weeks or longer improved blood oxygen saturation, while single-dose administration had minimal effect on oxygenation. The included studies were of moderate methodological quality and limited to Chinese settings [11].

Ruan et al. (2019) compared 30 mg versus 60 mg intravenous fasudil in 60 patients with congenital heart disease and severe pulmonary arterial hypertension. The 60 mg dose produced significantly greater reductions than 30 mg across all hemodynamic parameters: systolic pulmonary artery pressure (negative 13.1 versus negative 9.3 percent), mean pulmonary artery pressure (negative 12.4 versus negative 8.5 percent), and pulmonary vascular resistance (negative 35.8 versus negative 22.2 percent), all P less than 0.05 [12]. No systemic hypotension or serious adverse reactions were observed at either dose [12].

7.3 Amyotrophic lateral sclerosis

The ROCK-ALS trial (Koch et al. 2024) was a phase 2, randomized, double-blind, placebo-controlled trial at 19 centers in Germany, France, and Switzerland (NCT03792490) [13]. A total of 120 patients with ALS were randomized to fasudil 15 mg twice daily (30 mg group, n equals 35), fasudil 30 mg twice daily (60 mg group, n equals 39), or placebo (n equals 44), administered as intravenous infusion over 45 minutes for 20 consecutive working days, with mandatory riluzole co-medication.

The primary endpoint (safety) was met: safety probability was 1.00 in all groups, with no drug-related serious adverse events [13]. ALSFRS-R change from baseline to day 180 showed decline of negative 6.5 points (placebo), negative 7.94 points (30 mg), and negative 6.85 points (60 mg), with no significant differences between groups (P equals 0.23 for 30 mg, P equals 0.77 for 60 mg versus placebo at day 180) [13].

The electrophysiological biomarker MUNIX Megascor 10 (motor unit number index) showed significant dose-dependent preservation of motor units: at day 26, the fasudil 60 mg group showed a ratio of 1.01 versus 0.94 for placebo (P equals 0.015); at day 90, both fasudil groups showed 0.89 versus 0.79 for placebo (P equals 0.042 for 30 mg, P equals 0.038 for 60 mg) [13]. These differences were not sustained at day 180, approximately 160 days after treatment cessation. Serum glial fibrillary acidic protein (GFAP) was significantly reduced at day 180 in the 60 mg group (ratio 0.75, 95 percent CI 0.60 to 0.95; P equals 0.041). A prespecified subgroup analysis found significantly slower vital capacity decline in females in the 60 mg group at all timepoints [13].

CSF hydroxyfasudil concentrations confirmed central nervous system penetration: 5.8 plus or minus 2.0 micrograms per liter (30 mg) and 9.4 plus or minus 4.5 micrograms per liter (60 mg) [13].

The initial compassionate use study (Koch et al. 2020) treated three ALS patients with 30 mg fasudil intravenous twice daily for 20 working days. All three tolerated fasudil without adverse events; ALSFRS-R remained stable during treatment. One patient showed slow vital capacity improvement from 800 to 1,850 milliliters, and another showed MUNIX increase in 5 of 6 examined muscles [14].

7.4 Coronary vasospasm

Masumoto et al. (2002) demonstrated that intracoronary fasudil markedly attenuated acetylcholine-induced coronary vasoconstriction and prevented chest pain and ischemic electrocardiographic changes in all patients with vasospastic angina [31]. A subsequent study reported that the vasodilatory effect of fasudil at concentric coronary stenosis significantly exceeded that of nitroglycerin in patients with stable angina pectoris [38]. These findings established a mechanistic rationale for ROCK-mediated coronary vasospasm but have not progressed to a registration program.

7.5 Combination therapy

In a retrospective study of 104 patients with subarachnoid hemorrhage complicated by cerebral vasospasm, the combination of butylphthalide with fasudil hydrochloride (30 mg three times daily for two weeks) produced significant improvements in inflammatory markers (NSE, TNF-alpha, IL-8, CRP), cognitive function scores, and vascular endothelial function compared to fasudil alone [28].

8. Sourcing and Quality Verification

Research-grade fasudil hydrochloride is widely available from multiple established chemical suppliers including Tocris (catalog 0541), Sigma-Aldrich, Cayman Chemical, MedChemExpress (HY-10341A), Selleckchem, and others at greater than 98 percent purity by high-performance liquid chromatography. Supply is available in milligram to gram quantities as the hydrochloride salt.

Pharmaceutical-grade fasudil hydrochloride hydrate (Eril; Asahi Kasei Pharma) is marketed in Japan as a 30 mg lyophilized preparation for reconstitution for intravenous administration. In the compassionate use ALS study, Eril ampoules were used directly for clinical research [14]. Pharmaceutical sourcing is feasible for investigators in Japan and through international pharmaceutical import channels for jurisdictions permitting named-patient imports.

Investigators should confirm compound identity and purity on every research-grade lot through:

- High-performance liquid chromatography area-percent purity (target greater than 98 percent)
- Mass spectrometry confirmation of molecular weight (protonated molecular ion $[M+H]^+$ at m/z 292.11, consistent with the monoisotopic mass of 291.104 g/mol)
- Nuclear magnetic resonance spectroscopy for structural identity confirmation (characteristic isoquinoline aromatic proton signals and homopiperazine CH₂ resonances)
- Residual solvent analysis for common synthesis solvents

The compound is achiral; enantiomeric purity verification is not required. Investigators conducting sensitive dose-response studies should verify the hydration state and salt stoichiometry of each lot, as lot-to-lot variability in the hydrochloride hydrate stoichiometry may affect dosing precision. Fasudil is catalogued in PubChem as CID 3547 (SMILES: C1CNCCN(C1)S(=O)(=O)C2=CC=CC3=C2C=CN=C3, InChIKey: NGOGFTYYXHNFQH-UHFFFAOYSA-N), in ChEMBL as ChEMBL38380, and in DrugBank as DB08162; these identifiers enable unambiguous lot verification against reference databases.

For in vitro assays, investigators should note that fasudil's cell-based IC₅₀ (1.9 to 10.7 micromolar depending on isoform) requires working concentrations higher than those used for more potent ROCK inhibitors such as Y-27632 or ripasudil, and stock solutions should be prepared at concentrations compatible with the target working range.

9. Reconstitution and Handling

Fasudil hydrochloride is freely soluble in water at room temperature. The pharmaceutical-grade preparation (Eril) is supplied as a lyophilized powder in glass vials, reconstituted in 0.9 percent normal saline. For clinical and clinical-research intravenous applications, the reconstituted solution is diluted in 50 to 100 milliliters of normal saline and administered as an infusion over 30 to 45 minutes [4, 13, 14].

For in vitro research, stock solutions are typically prepared at 10 to 100 millimolar in water, dimethyl sulfoxide (DMSO), or normal saline. Aqueous stock solutions are stable at 2 to 8 degrees Celsius for at least 14 days. DMSO stocks at concentrated preparations should be aliquoted to minimize freeze-thaw cycles. The compound is stable at room temperature in dry conditions as a solid; refrigerated storage (2 to 8 degrees Celsius) is recommended for long-term storage of both solid and reconstituted forms. Light protection is not specifically required but is reasonable practice.

For in vivo animal research, fasudil is administered by intraperitoneal injection (dissolved in normal saline, typical doses 3 to 30 mg/kg), oral gavage (in water or saline, 30 to 100 mg/kg), or in drinking water (30 to 100 mg/kg/day calculated from estimated daily water intake) [19, 21, 24]. The oral route achieves pharmacologically relevant hydroxyfasudil concentrations despite near-complete first-pass metabolism of the parent compound [15]. Inhaled delivery via liposomal formulation has been explored in preclinical studies, producing prolonged pulmonary preferential vasodilation with a pulmonary half-life of 1.17 plus or minus 0.21 hours (compared to 0.39 plus or minus 0.12 hours for intravenous in rats) and pulmonary bioavailability of approximately 41 percent [39].

10. Stack Interaction Considerations

Fasudil interacts with other compounds principally through pharmacodynamic mechanisms (shared downstream pathways) rather than through conventional metabolic drug-drug interactions. The aldehyde oxidase-dependent metabolism, rather than cytochrome P450-dependent, reduces the risk of conventional CYP-mediated interactions.

Metabolic interactions: Fasudil is metabolized by aldehyde oxidase, not by CYP enzymes [16]. Standard CYP inhibitors and inducers (ketoconazole, rifampin, carbamazepine) are not expected to significantly alter fasudil or hydroxyfasudil exposure. However, aldehyde oxidase inhibitors (raloxifene, menadione, certain flavonoids) could theoretically increase fasudil exposure and decrease hydroxyfasudil formation, though this interaction has not been characterized clinically. The species-specific metabolic pathway means that compounds altering aldehyde oxidase activity may have effects in humans not predictable from rat or dog models [16].

Other ROCK inhibitors: Concurrent administration with Y-27632, ripasudil, or other ROCK inhibitors produces additive ROCK inhibition. The combination is pharmacologically redundant for systemic targets and has not been characterized for safety. In stem cell culture, fasudil and Y-27632 are used interchangeably but not in combination [34].

Calcium channel blockers: Concurrent use with nimodipine has been studied in clinical trials without reported adverse interaction; the two agents act through different mechanisms (ROCK inhibition versus calcium channel blockade), and their vasodilatory effects may be additive [5, 6]. Blood pressure monitoring is appropriate in combination use.

Antihypertensive agents: The vasodilatory mechanism of fasudil may produce additive blood pressure reduction when combined with antihypertensive drugs. While clinically significant hypotension has not been a prominent finding in fasudil trials, investigators conducting research with concurrent antihypertensives should monitor for additive effects [9, 15].

Vasospasm prophylaxis combinations: Japanese clinical practice commonly combines fasudil with cilostazol and/or statins for vasospasm prophylaxis after subarachnoid hemorrhage. The cilostazol-fasudil-statin triple combination was the most effective prophylactic regimen in a nationwide survey (odds ratio 0.38 for delayed cerebral infarction) [29].

Anticoagulants and antiplatelet agents: Fasudil's ROCK inhibition may modestly affect platelet function through ROCK-dependent platelet activation pathways. The postmarketing surveillance of 1,462 patients did not report excess hemorrhagic events [7].

Riluzole: In the ROCK-ALS trial, mandatory riluzole co-medication (50 mg twice daily) was administered concurrently with fasudil without apparent interaction or excess adverse events [13]. Riluzole is metabolized primarily by CYP1A2, which does not interact with the aldehyde oxidase pathway.

Corticosteroids: Concurrent use would produce additive anti-inflammatory effects through different mechanisms (NF-kappaB suppression by fasudil via ROCK, glucocorticoid receptor-mediated suppression by corticosteroids). No specific safety concern has been identified.

11. Adverse Events and Safety

11.1 Common adverse events

The adverse event profile of fasudil has been characterized across multiple clinical settings, from the pivotal cerebral vasospasm trial (267 patients) to the postmarketing surveillance (1,462 patients) and the ROCK-ALS Phase 2 trial (120 patients).

In the SAFE-ROCK Phase I trial in 14 healthy volunteers, no serious adverse events occurred. One subject (7.1 percent) reported headache with intravenous treatment (assessed as drug-related) and one subject reported fatigue with oral treatment (assessed as not drug-related). A statistically significant but clinically mild increase in indigestion symptoms was observed on the Gastrointestinal Symptom Rating Scale (P equals 0.048) [15].

In the ROCK-ALS trial, 352 adverse events occurred in 92 of 118 (78.0 percent) patients across all groups, consistent with the high background event rate in ALS. Of these, 58 percent were grade 1, 36 percent were grade 2, and 6 percent were grade 3. Forty serious adverse events occurred in 30 of 120 (25.4 percent) patients; none were attributed to the study drug. No clinically significant changes in arterial blood pressure or heart rate were observed [13].

The overall adverse event profile across the clinical literature includes: mild systolic blood pressure reduction (typically 5 to 10 mmHg, generally asymptomatic), headache (reported in healthy volunteer and clinical trial settings), allergic skin reactions (postmarketing surveillance), mild gastrointestinal symptoms with oral dosing, and reversible renal impairment (rare, postmarketing) [7, 13, 15].

11.2 Safety in cerebral vasospasm

In the pivotal Shibuya et al. trial, no serious adverse events were reported across 14 days of thrice-daily intravenous dosing in 131 patients [4]. The postmarketing surveillance of 1,462 patients confirmed this safety profile: adverse event rates, including intracranial bleeding and hypotension, were similar to those observed in the

phase 3 trial [7]. In the fasudil versus nimodipine comparisons, adverse reaction rates were similar between groups (13 of 37 fasudil versus 15 of 35 nimodipine) with no serious adverse events in the fasudil groups [5, 6].

11.3 Safety in pulmonary hypertension

In patients with severe pulmonary hypertension, fasudil demonstrated selective pulmonary vasodilation without systemic hypotension [9]. A meta-analysis of 865 patients reported a pooled minor adverse event rate of 5.2 percent in fasudil groups versus 4.8 percent in controls, with no serious adverse events [11]. At both 30 mg and 60 mg doses in congenital heart disease patients, no systemic hypotension or serious adverse reactions occurred [12].

11.4 Hypotension considerations

Clinically significant systemic hypotension has not been a prominent feature of the safety profile. The pulmonary selectivity observed in hypertension studies (17 percent PVR reduction without systemic blood pressure change) [9] and the absence of significant blood pressure changes in the ROCK-ALS and SAFE-ROCK trials [13, 15] suggest that the degree of systemic vasodilation at therapeutic doses is modest. However, transient hypotension following intracoronary fasudil has been reported in approximately 20 percent of patients in coronary vasospasm studies, resolving without intervention [31].

11.5 Hemorrhagic considerations

Intracranial hemorrhage following intra-arterial fasudil administration for cerebral vasospasm has been reported as a rare adverse event. The incidence in post-marketing surveillance was similar to background rates of post-procedural hemorrhage [7]. Investigators using fasudil in the post-neurosurgical setting should maintain standard hemorrhagic monitoring.

11.6 Long-term safety

Long-term safety data are limited. The longest published treatment duration is 20 working days of intravenous administration (ROCK-ALS, with follow-up to day 180) [13]. Chronic oral dosing safety has not been characterized in published human studies. The theoretical concern of chronic ROCK inhibition relates to the role of

ROCK in tissue homeostasis, wound healing, and immune function; these concerns have not materialized in the available clinical data, though the treatment durations studied are relatively short.

11.7 Hepatic and renal considerations

Hepatic impairment may alter fasudil metabolism through reduced aldehyde oxidase activity. No formal hepatic impairment pharmacokinetic study has been published. Reversible renal impairment has been reported rarely in postmarketing surveillance; the mechanism is not characterized [7]. No human pregnancy data are available; use in pregnant subjects is not recommended outside formal investigational settings.

12. Comparable Compounds Assessment

This section evaluates five compounds in the ROCK inhibitor and cerebral vasospasm pharmacological space against fasudil on five competency standards: novelty, effect size, promising potential, side-effect profile, and overall validation. Each standard is scored 1 to 5.

12.1 Y-27632

Mechanism: Selective ROCK inhibitor with slightly preferential activity at ROCK1 (Ki 140 to 220 nanomolar) versus ROCK2 (Ki approximately 300 nanomolar). The compound features a 4-aminopyridine core structurally unrelated to fasudil's isoquinolinesulfonamide scaffold [35].

Development trajectory: Y-27632 was characterized in the landmark Uehata et al. (1997) Nature publication establishing the role of ROCK in calcium sensitization and hypertension [3]. The compound has not undergone clinical development and is not approved for any indication. It is the most widely used ROCK inhibitor in basic research, particularly in stem cell biology.

Novelty: 3/5. The first well-characterized selective ROCK inhibitor but predates the clinical development era.

Effect size: Not applicable. No clinical data. Preclinical activity is broadly comparable to fasudil.

Promising potential: 2/5. No clinical development planned; serves as a reference research standard.

Side-effect profile: 3/5. Preclinical only; no human safety data.

Overall validation: 2/5. No clinical validation despite extensive preclinical characterization.

Comparison to fasudil: Y-27632 has slightly greater ROCK selectivity (non-selective against 4 of 25 kinases versus 8 of 27 for fasudil) but requires a seven-step synthesis (45 percent yield) and is 10 to 30 times more expensive than fasudil [34]. Fasudil has the decisive advantages of clinical approval, human pharmacokinetic data, and a safety database spanning thousands of patients.

12.2 Ripasudil (K-115, Glanatec)

Mechanism: ROCK inhibitor structurally derived from fasudil (fluorinated analog) with IC₅₀ values of 19 nanomolar (ROCK1) and 51 nanomolar (ROCK2), indicating slightly higher ROCK1 potency [35]. Optimized for topical ophthalmic tissue penetration.

Development trajectory: Developed by Kowa Co., Ltd. (Aichi, Japan). Approved in Japan in 2014 as the first small-molecule selective ROCK inhibitor marketed for glaucoma and ocular hypertension (0.4 percent ophthalmic solution, twice daily). Also approved in China [36].

Novelty: 3/5. Meaningful advance in ophthalmic ROCK inhibitor design but restricted to topical delivery.

Effect size: Intraocular pressure reduction of 2.0 to 4.8 mmHg; higher baseline IOP (greater than 21 mmHg) yielded reductions up to 4.8 mmHg at peak [36].

Promising potential: 3/5. Established in its ophthalmic niche. No systemic development.

Side-effect profile: 3/5. Conjunctival hyperemia is the principal adverse event (4 percent in 2-year postmarketing surveillance), with conjunctival hemorrhage and blepharitis also reported [36].

Overall validation: 3/5. Approved and marketed with established safety.

Comparison to fasudil: Ripasudil and fasudil share the isoquinolinesulfonamide scaffold, with ripasudil optimized for topical ocular delivery and fasudil for systemic intravenous use. The two compounds serve complementary, non-overlapping clinical roles.

12.3 Netarsudil (Rhopressa)

Mechanism: Dual ROCK inhibitor and norepinephrine transporter (NET) inhibitor. K_i approximately 1 nanomolar at both ROCK1 and ROCK2, the highest potency among approved ROCK inhibitors [35]. The NET inhibition contributes to aqueous humor outflow through adrenergic mechanisms.

Development trajectory: Developed by Aerie Pharmaceuticals. FDA-approved in 2017 (0.02 percent ophthalmic solution) for glaucoma and ocular hypertension. Approved in the European Union in 2021 [36].

Novelty: 4/5. The dual ROCK/NET mechanism is a pharmacological innovation among approved ROCK inhibitors.

Effect size: IOP reduction from baseline of 21.9 to 23.7 mmHg to 17.5 to 19.5 mmHg; non-inferior to timolol 0.5 percent twice daily [36].

Promising potential: 3/5. Established in ophthalmic indication. No systemic development.

Side-effect profile: 2/5. Conjunctival hyperemia (54.4 percent), cornea verticillata (20.9 percent), and conjunctival hemorrhage (17.2 percent) are common [36].

Overall validation: 4/5. FDA-approved with robust clinical trial data.

Comparison to fasudil: Netarsudil is the most potent ROCK inhibitor at the molecular level (K_i approximately 1 nanomolar versus K_i 47 to 76 nanomolar for fasudil) but is restricted to topical ophthalmic delivery. The systemic pharmacology and clinical indications do not overlap with fasudil's cerebrovascular and pulmonary applications.

12.4 Belumosudil (KD025, Rezurock)

Mechanism: Selective ROCK2 inhibitor (IC_{50} approximately 153 nanomolar for ROCK2) [37]. The ROCK2 selectivity distinguishes belumosudil from all other approved ROCK inhibitors. ROCK2 inhibition restores the Th17/regulatory T cell balance toward regulatory T cells via a STAT5-dependent mechanism and reduces STAT3-driven proinflammatory signaling [37].

Development trajectory: Developed by Kadmon (now Sarepta Therapeutics). FDA-approved in July 2021 for chronic graft-versus-host disease (cGvHD) in patients aged 12 and older after failure of two or more prior systemic therapies. First-in-class designation [37].

Novelty: 5/5. ROCK2-selective mechanism and the cGvHD indication represent genuine pharmacological and therapeutic innovation.

Effect size: Phase IIa overall response rate of 65 percent (200 mg once daily), 69 percent (200 mg twice daily), and 62 percent (400 mg once daily) across 54 patients with refractory cGvHD. Median duration of response 35 weeks. Two-year overall survival 82 percent. Corticosteroid dose reduction in 67 percent of patients [37].

Promising potential: 4/5. FDA-approved and commercially available for a serious unmet need.

Side-effect profile: 3/5. Upper respiratory infection (46 percent), diarrhea (33 percent), fatigue (33 percent), nausea (33 percent). Low rates of grade 3 or higher cytopenias (4 percent) [37].

Overall validation: 5/5. FDA-approved with strong clinical evidence.

Comparison to fasudil: Belumosudil and fasudil represent different poles of the ROCK inhibitor spectrum: belumosudil is ROCK2-selective and orally bioavailable for an immunological indication; fasudil is a pan-ROCK inhibitor administered intravenously for vascular and neurological indications. Belumosudil demonstrates that selective ROCK2 inhibition can be clinically effective, informing the understanding of fasudil's ROCK2-preferring pharmacology. The two compounds do not compete for clinical indications but are complementary in the broader ROCK biology landscape.

12.5 Nimodipine

Mechanism: Dihydropyridine calcium channel blocker (L-type) with preferential cerebrovascular activity. Nimodipine directly blocks voltage-gated calcium channels, reducing calcium-dependent contraction through a mechanism entirely distinct from the ROCK-mediated calcium sensitization pathway inhibited by fasudil.

Development trajectory: Approved in multiple jurisdictions for prevention and treatment of cerebral vasospasm following subarachnoid hemorrhage. Nimodipine is the international standard of care for this indication and the principal active comparator against which fasudil has been evaluated [5, 6].

Novelty: 2/5. Well-established mechanism, non-novel within the vasospasm pharmacology space.

Effect size: Moderate reductions in cerebral vasospasm and clinical outcome improvement after SAH. In the fasudil comparison, good clinical outcome was 61.7 percent with nimodipine versus 74.5 percent with fasudil (P equals 0.040) [6].

Promising potential: 3/5. Established and widely available but limited by mechanism (does not address the ROCK-mediated component of vasospasm).

Side-effect profile: 3/5. Systemic hypotension is the principal adverse effect, particularly with intravenous formulations.

Overall validation: 4/5. International standard of care with decades of clinical use.

Comparison to fasudil: Nimodipine and fasudil address different components of vasospasm pathophysiology: nimodipine targets calcium-dependent contraction while fasudil targets ROCK-mediated calcium sensitization. Head-to-head trials suggest fasudil produces comparable or superior clinical outcomes (74.5 versus 61.7 percent) with a similar or favorable safety profile [6]. Japanese clinical practice has adopted fasudil as the standard agent; in Western jurisdictions where fasudil is not approved, nimodipine remains the standard of care.

12.6 Comparator summary

The five comparators occupy distinct positions. **Y-27632** is the research standard for ROCK inhibition but lacks clinical development. **Ripasudil** and **netarsudil** are ophthalmic-specific ROCK inhibitors with no systemic application overlap. **Belumosudil** is the most innovative comparator, demonstrating that ROCK2-selective inhibition achieves FDA approval for an immunological indication. **Nimodipine** is the most clinically relevant comparator, representing the established standard of care against which fasudil has demonstrated comparable or superior efficacy.

Fasudil occupies a unique position as the first and only systemically administered ROCK inhibitor approved for clinical use, with three decades of safety data, expanding investigational applications, confirmed CNS penetration of the active metabolite, and a favorable cost-to-potency ratio for both clinical and research applications. Its principal limitations are the absence of an approved oral formulation, restriction of marketing approval to Japan and China, and moderate kinase selectivity relative to newer agents. Its principal strengths are the established clinical safety database, the dual vascular and neuroprotective pharmacology, the comprehensive pharmacokinetic characterization, and the confirmed positioning as standard of care in Japanese neurosurgical practice.

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