

RESEARCH MONOGRAPH . COMPOUND PROFILE

R-13

A rationally designed bis-carbamate prodrug of 7,8-dihydroxyflavone engineered for improved oral bioavailability and sustained brain-derived neurotrophic factor receptor activation, advancing through Phase 1 clinical evaluation for Alzheimer's disease and under preclinical investigation for amyotrophic lateral sclerosis, depression, Parkinson's disease, and peripheral nerve regeneration.

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ABSTRACT

R-13 (BrAD-R13, Braegen-01) is a synthetic bis-methylcarbamate prodrug of 7,8-dihydroxyflavone (7,8-DHF; tropoflavin), designed to overcome the poor oral bioavailability and rapid hepatic conjugation of the parent flavone while preserving its selective agonist activity at the tropomyosin receptor kinase B (TrkB) receptor, the principal high-affinity signaling receptor for brain-derived neurotrophic factor (BDNF). The compound was identified through systematic medicinal chemistry optimization of ester and carbamate modifications on the 7,8-DHF catechol ring by Chen, Ye, and colleagues at Emory University School of Medicine and Zhejiang University, with R-13 selected from among twenty candidate derivatives as the sole compound satisfying all screening criteria for gastric acid stability, intestinal absorption, hydrolyzability in liver microsomes and plasma, and adequate membrane permeability. Following oral administration, R-13 is hydrolyzed through a monophenol intermediate (designated T1) to release 7,8-DHF, which binds the extracellular domain of TrkB and triggers receptor dimerization, autophosphorylation, and activation of the downstream Akt and ERK/MAPK signaling cascades. In mice, R-13 at 36 mg/kg oral dose produces a maximum plasma concentration of 129 ng/mL at 30 minutes, with an elimination half-life of approximately 220 minutes and oral bioavailability of 10.5 percent (compared to 4.6 percent for the parent 7,8-DHF), and sustains brain 7,8-DHF concentrations above 8 ng/g for at least 4 hours. The pharmacological consequences of sustained TrkB activation are broad: R-13 represses asparagine endopeptidase (AEP, also termed delta-secretase), a protease implicated in the pathological cleavage of

both amyloid precursor protein and tau in Alzheimer's disease, thereby reducing amyloid-beta deposition and neurofibrillary tangle formation. Chronic oral administration of R-13 in the 5XFAD transgenic mouse model of Alzheimer's disease dose-dependently restored dendritic spine density, enhanced long-term potentiation, reduced senile plaque burden, attenuated neuroinflammatory cytokine levels (interleukin-1-beta, interleukin-6, tumor necrosis factor alpha), and reversed spatial and working memory deficits in the Morris water maze without demonstrable toxicity at 12 weeks of continuous dosing at up to 43.6 mg/kg/day. Additional preclinical evidence supports efficacy in the SOD1-G93A transgenic mouse model of amyotrophic lateral sclerosis, where R-13 preserved motor neuron counts, reduced gastrocnemius muscle atrophy, attenuated glial activation, and enhanced mitochondrial biogenesis through AMPK/PGC-1-alpha/Nrf1/Tfam pathway activation. In a peripheral nerve transection model, oral R-13 produced axon regeneration and functional electromyographic recovery superior to the parent 7,8-DHF. R-13 has also been characterized as preventing ovariectomy-induced bone loss through TrkB/Akt-mediated inhibition of AEP and upregulation of osteoprotegerin. Braegen Pharmaceutical (Shenzhen, China) has completed a Phase 1 clinical trial of BrAD-R13 in Alzheimer's disease, the first clinical evaluation of a TrkB agonist prodrug in this indication, with plans for Phase 2 efficacy trials. The compound is not approved in any jurisdiction for any indication. This monograph reviews the chemistry, synthesis, and prodrug design of R-13; the TrkB receptor pharmacology and downstream signaling; the comprehensive preclinical pharmacokinetic profile; the evidence base across Alzheimer's disease, amyotrophic lateral sclerosis, peripheral nerve injury, bone metabolism, and neuropsychiatric indications; sourcing and

handling for research applications; stack-interaction considerations; adverse-event signal; and a comparative assessment of five alternative TrkB agonist and BDNF-mimetic candidates against R-13 on five competency standards.

1. Compound Identification

1.1 Names, synonyms, and registry identifiers

Nonproprietary designation: R-13 (also written R13). The compound does not have an International Nonproprietary Name assigned by the World Health Organization.

Development codes: BrAD-R13 (Braegen Pharmaceutical), Braegen-01 (company designation for clinical development). The Ye laboratory at Emory University and Zhejiang University uses the designation R-13 in primary publications [1].

Chemical Abstracts Service registry number: 1609067-49-3.

PubChem compound identifier: 90117609.

Unique Ingredient Identifier (UNII): OGG19T0LU2.

International Union of Pure and Applied Chemistry name: [8-(methylcarbamoyloxy)-4-oxo-2-phenylchromen-7-yl] N-methylcarbamate [2].

1.2 Structural class and chemistry

R-13 is a synthetic flavone derivative in which both hydroxyl groups of the catechol ring (positions 7 and 8) of 7,8-dihydroxyflavone are masked as methylcarbamate esters. The structural class places R-13 within the flavonoid family (2-phenylchromen-4-one core), modified by the introduction of carbamate prodrug moieties designed to protect the catechol from rapid phase II conjugation (glucuronidation, sulfation, and methylation) in the liver [1, 3].

Molecular formula: C₁₉H₁₆N₂O₆.

Molecular weight: 368.345 g/mol (monoisotopic mass 368.101 g/mol).

Physical properties: Off-white to pale yellow solid. Soluble in dimethyl sulfoxide; sparingly soluble in aqueous media without organic cosolvent. The compound is less water-soluble than the parent 7,8-DHF owing to the lipophilic carbamate groups.

Parent compound: 7,8-dihydroxyflavone (7,8-DHF; tropoflavin; CAS 38183-03-8; molecular formula C₁₅H₁₀O₄; molecular weight 254.24 g/mol). The parent compound is a naturally occurring flavone first isolated from plant sources and subsequently identified as a selective TrkB agonist by Jang et al. (2010) [4].

Structural relationship to parent: R-13 is a bis-methylcarbamate derivative in which each catechol hydroxyl group carries an N-methylcarbamate group (forming two carbamate ester bonds). The 2-phenyl group, the chromen-4-one core, and the overall flavone geometry are preserved. Upon hydrolysis in liver microsomes and plasma, R-13 releases the free catechol 7,8-DHF through a monophenol intermediate designated T1 [1].

1.3 Synthesis

The published synthesis of R-13 proceeds from commercially available 7,8-dihydroxyflavone. The two catechol hydroxyl groups are converted to methylcarbamate esters by reaction with methyl isocyanate under anhydrous conditions in the presence of a suitable base catalyst. Chen et al. (2018) reported the synthesis of approximately twenty ester and carbamate derivatives of 7,8-DHF as part of a systematic prodrug optimization campaign [1]. Of these, six candidates (designated R5, R7, R13, R16, R17, and R18) met initial stability criteria including resistance to gastric acid hydrolysis (pH 1.2), acceptable intestinal absorption, and sufficient membrane permeability. R-13 was the sole candidate that additionally demonstrated ready hydrolyzability in both mouse and human liver microsomes, distinguishing it from R7, which showed minimal conversion in human liver tissue [1, 2]. The product is purified by silica gel chromatography and characterized by nuclear magnetic resonance spectroscopy, mass spectrometry, and high-performance liquid chromatography. Purity of research-grade material is typically greater than 95 percent by HPLC [5, 6].

2. Abstract

R-13 (BrAD-R13, Braegen-01) is an orally bioavailable bis-methylcarbamate prodrug of 7,8-dihydroxyflavone (7,8-DHF), a selective small-molecule agonist of the tropomyosin receptor kinase B (TrkB) receptor [1, 4]. The compound was developed to overcome the principal translational limitation of 7,8-DHF: poor oral bioavailability (approximately 4.6 percent in mice) resulting from rapid hepatic conjugation of the catechol ring by glucuronidation, sulfation, and O-methylation [3, 7]. R-13 increases oral bioavailability of the active 7,8-DHF moiety to approximately 10.5 percent, extends the plasma elimination half-life from 134 minutes to approximately 220 minutes, and produces sustained brain concentrations of free 7,8-DHF for at least 4 hours after a single oral dose [1].

The mechanism of action is indirect: R-13 itself does not bind TrkB but serves as a depot for the controlled release of 7,8-DHF, which binds the extracellular leucine-rich motif and cysteine cluster 2 (LRM/CC2) domain of TrkB with high selectivity over the related TrkA and TrkC receptors [4, 8]. Receptor binding provokes dimerization, autophosphorylation at tyrosine 705 and 706, and activation of the Akt and ERK/MAPK signaling cascades that recapitulate the neurotrophic and neuroprotective effects of BDNF [1, 4]. A downstream consequence of particular relevance to Alzheimer's disease is the Akt-mediated phosphorylation and inactivation of asparagine endopeptidase (AEP, also called delta-secretase or legumain), a cysteine protease that cleaves both amyloid precursor protein and tau at specific asparagine residues, generating neurotoxic fragments that drive amyloid plaque deposition and neurofibrillary tangle formation [9, 10].

Chronic oral administration of R-13 in the 5XFAD transgenic mouse model of Alzheimer's disease (which expresses five familial Alzheimer mutations and develops aggressive amyloid pathology) produced dose-dependent reductions in amyloid-beta 40 concentration, senile plaque deposition, hippocampal synapse loss, and neuroinflammatory cytokine levels, and rescued spatial memory deficits in the Morris water maze at doses of 7.25 to 43.6 mg/kg/day over 3 months [1]. A 12-week chronic toxicity study in wild-type C57BL/6 mice at the highest dose (43.6 mg/kg/day) demonstrated no detectable organ toxicity [1]. Preclinical evidence has been extended to amyotrophic lateral sclerosis (SOD1-G93A transgenic mice), peripheral nerve injury (sciatic nerve transection), ovariectomy-induced osteoporosis

sis, and models of depression and Parkinson's disease, with consistent demonstrations of TrkB-dependent neuroprotective, neurotrophic, and anti-inflammatory effects [11, 12, 13, 14].

Braegen Pharmaceutical completed a Phase 1 clinical trial of BrAD-R13 in healthy volunteers and Alzheimer's disease patients, the first human evaluation of a TrkB agonist prodrug for this indication [15]. Early results indicate tolerability as an oral tablet formulation, with plans for Phase 2 efficacy evaluation [15]. The compound is not approved in any jurisdiction. It is available as a research-grade preparation from multiple chemical suppliers; investigators should confirm identity and purity on every lot.

3. Discovery and Development History

3.1 Discovery of 7,8-dihydroxyflavone as a TrkB agonist

The discovery history of R-13 begins with the identification of its parent compound, 7,8-dihydroxyflavone (7,8-DHF), as a selective small-molecule TrkB agonist. Jang et al. (2010) at Emory University reported a cell-based high-throughput screen of a library of small molecules for compounds that activate TrkB receptor signaling in cultured neurons [4]. 7,8-DHF emerged as a potent hit that provoked TrkB dimerization and autophosphorylation, activated the downstream Akt and ERK/MAPK cascades, and promoted neuronal survival in a TrkB-dependent manner. The compound crossed the blood-brain barrier after intraperitoneal injection, inhibited kainic acid-induced excitotoxicity, reduced infarct volumes in a mouse stroke model, and was neuroprotective in a mouse model of Parkinson's disease [4]. The Jang et al. report was the first identification of a small-molecule TrkB agonist and established 7,8-DHF as the foundational tool compound for the field.

3.2 Limitation of the parent compound

Despite robust preclinical activity in models of neurodegeneration, stroke, depression, traumatic brain injury, and metabolic disease, 7,8-DHF was limited by poor oral bioavailability (approximately 4.6 percent in mice) arising from rapid hepatic phase II metabolism of the catechol ring [3, 7]. Glucuronidation and sulfation of the 7- and 8-hydroxyl groups, together with catechol-O-methyltransferase-mediated methylation, produced extensive first-pass clearance. Both O-methylated metabolites (8-hydroxy-7-methoxyflavone and 7-hydroxy-8-methoxyflavone) retained some TrkB activity in vitro but had substantially reduced potency [3]. The clinical translation of 7,8-DHF was therefore contingent on either a formulation strategy to improve bioavailability or a prodrug strategy to protect the catechol.

3.3 Prodrug optimization and selection of R-13

Chen et al. (2018) at Emory University and Zhejiang University undertook a systematic prodrug campaign in which approximately twenty ester and carbamate derivatives of 7,8-DHF were synthesized and screened against a multiparameter optimiza-

tion panel [1]. The panel evaluated gastric acid stability (pH 1.2), intestinal permeability (Caco-2 monolayer), hydrolysis kinetics in mouse and human liver microsomes, and hydrolysis in mouse and human plasma. Six candidates (R5, R7, R13, R16, R17, R18) passed initial stability criteria; of these, R-13 (the bis-N-methylcarbamate) was the only compound that satisfied all screening requirements, including the critical demonstration of ready hydrolysis in human liver microsomes [1]. This species-specific hydrolysis criterion was essential because a predecessor compound, R7, showed adequate activation in mouse liver but minimal conversion in human liver tissue, which would have precluded clinical translation [1, 2].

3.4 Preclinical development

Between 2018 and 2022, the research program expanded from the initial Alzheimer's disease demonstration to multiple preclinical indications. Li et al. (2021) characterized R-13 in the SOD1-G93A transgenic mouse model of amyotrophic lateral sclerosis [11]. English et al. (2022) demonstrated enhanced axon regeneration and functional recovery after sciatic nerve transection [12]. Xiong et al. (2022) reported prevention of ovariectomy-induced bone loss through TrkB/Akt-mediated AEP inhibition and osteoprotegerin upregulation [13]. An optimized second-generation derivative, CF3CN, was also characterized by Chen et al. (2021) with further improved druglike properties [16], though R-13 remained the lead clinical candidate.

3.5 Clinical translation

Braegen Pharmaceutical, founded to advance the Ye laboratory compound portfolio, relocated its headquarters from Shanghai to Shenzhen's Brain Science and Technology Innovation Center in 2024 [15]. The company completed a Phase 1 clinical trial of BrAD-R13 (the clinical designation for R-13) formulated as an oral tablet, the first clinical evaluation of a TrkB agonist prodrug in Alzheimer's disease [15]. Early results were reported as demonstrating tolerability with potential signals of brain cell protection and cognitive benefit, though detailed clinical pharmacokinetic and safety data from the Phase 1 trial have not been published in peer-reviewed form as of the most recent monograph revision. Phase 2 efficacy trials are planned.

4. Mechanism of Action

R-13 is a prodrug; its pharmacological activity derives entirely from the released active metabolite, 7,8-dihydroxyflavone (7,8-DHF), which acts as a selective agonist at the tropomyosin receptor kinase B (TrkB) receptor. The mechanism is therefore a two-stage process: prodrug activation (hydrolysis to 7,8-DHF) followed by receptor-mediated signaling.

4.1 Prodrug activation

Following oral absorption, R-13 is hydrolyzed in the liver by hepatic esterases and carboxylesterases. The hydrolysis proceeds through a monophenol intermediate (T1, in which one carbamate group has been cleaved) to yield free 7,8-DHF [1]. The carbamate linkage confers gastric acid stability (R-13 is stable at pH 1.2) while permitting enzymatic cleavage in hepatic tissue. The rate of hydrolysis in mouse liver microsomes is rapid; in human liver microsomes, R-13 is likewise readily converted to 7,8-DHF, a critical translational property that distinguished it from the predecessor R7 [1, 2].

4.2 TrkB receptor agonism

TrkB (encoded by the NTRK2 gene) is the high-affinity signaling receptor for brain-derived neurotrophic factor (BDNF) and neurotrophin-4 (NT-4). It is a single-pass transmembrane receptor tyrosine kinase expressed at high density on neurons throughout the cortex, hippocampus, basal forebrain, substantia nigra, spinal motor neurons, and dorsal root ganglia, and at lower density on astrocytes, oligodendrocytes, and osteoblasts [17, 18].

7,8-DHF binds the extracellular domain of TrkB at the leucine-rich motif and cysteine cluster 2 (LRM/CC2) region, a site that overlaps partially with but is not identical to the BDNF binding site [4, 8]. Binding provokes receptor dimerization and autophosphorylation at tyrosine residues 705 and 706 in the kinase activation loop, initiating three principal downstream signaling cascades:

(a) **Akt/protein kinase B pathway.** Phosphorylation of Akt promotes neuronal survival through inhibition of pro-apoptotic factors (BAD, caspase-9) and activation of the mammalian target of rapamycin (mTOR) signaling complex, which supports protein synthesis, dendritic spine growth, and synaptic plasticity [17].

(b) **ERK/MAPK pathway.** Activation of the Ras-Raf-MEK-ERK cascade promotes cell proliferation, differentiation, and long-term potentiation in hippocampal circuits [1, 4].

(c) **PLC-gamma/calcium pathway.** Phospholipase C gamma activation generates inositol trisphosphate and diacylglycerol, raising intracellular calcium and activating calcium/calmodulin-dependent protein kinase II, which contributes to synaptic plasticity and CREB-mediated transcription [17].

7,8-DHF is selective for TrkB over the related neurotrophin receptors TrkA (the NGF receptor) and TrkC (the NT-3 receptor). It does not bind the low-affinity neurotrophin receptor p75NTR, an important distinction because p75NTR activation can promote apoptosis and growth cone collapse through opposing signaling mechanisms [4, 12].

4.3 Asparagine endopeptidase (AEP/delta-secretase) inhibition

A pharmacologically critical downstream effect of TrkB/Akt activation is the phosphorylation of asparagine endopeptidase (AEP, also known as delta-secretase or legumain) at threonine 322 [9, 10]. Phosphorylation at this site inactivates AEP, preventing its cysteine protease activity. AEP cleaves both amyloid precursor protein (APP) at asparagine 585 and tau at asparagine 368, generating neurotoxic fragments: the APP N585 fragment promotes amyloidogenic beta-secretase processing and amyloid-beta production, while the tau N368 fragment is hyperphosphorylation-prone and aggregation-competent [9, 10]. AEP activity increases with age and is markedly elevated in Alzheimer's disease brain tissue. R-13, through sustained TrkB/Akt activation, maintains AEP in a phosphorylated (inactive) state, simultaneously reducing both amyloid-beta deposition and tau pathology [1, 9].

4.4 Mitochondrial and metabolic effects

In the SOD1-G93A amyotrophic lateral sclerosis model, R-13 activated the AMPK/PGC-1-alpha/Nrf1/Tfam signaling cascade, promoting mitochondrial biogenesis, upregulating oxidative phosphorylation complex proteins (Cox5a, SDHB, ATP5a), increasing ATP production, and reducing lipid peroxidation (measured by malondialdehyde) in spinal cord and medulla oblongata [11]. These mitochondrial effects represent a downstream consequence of TrkB signaling through AMPK and are distinct from, though complementary to, the Akt-mediated AEP inhibition relevant to Alzheimer's disease.

4.5 CREB activation and osteoprotegerin upregulation

In bone metabolism, 7,8-DHF released from R-13 activates TrkB and its downstream effector CREB (cAMP response element-binding protein), which is critical for osteoprotegerin (OPG) augmentation [13]. Concurrently, the C/EBP-beta/AEP pathway is repressed, inhibiting RANKL-induced osteoclastogenesis. R-13 treatment elevated OPG without altering RANKL levels in ovariectomized mice, blocking trabecular bone loss with efficacy equivalent to anti-RANKL antibody therapy [13].

4.6 Selectivity considerations

The selectivity of the TrkB agonism mediated by R-13/7,8-DHF has been the subject of scientific debate. A 2017 report by Bhatt et al. using multiplex quantitative assays questioned whether 7,8-DHF directly activates TrkB, suggesting that some reported agonists may instead produce delayed TrkB transactivation through G-protein coupled receptor (GPCR) intermediaries [19]. However, the Chen et al. (2018) demonstration of TrkB autophosphorylation within 15 minutes of R-13 administration in vivo, the dose-dependent activation of downstream Akt and MAPK pathways, and the abolition of efficacy in TrkB-deficient systems support direct or functionally equivalent agonist activity [1, 12]. The mechanistic debate remains active but does not alter the observed therapeutic pharmacology in preclinical disease models.

5. Pharmacokinetics and Routes of Administration

5.1 Absorption

R-13 is absorbed from the gastrointestinal tract after oral administration. The compound is stable at gastric pH (pH 1.2), permitting transit through the stomach without premature hydrolysis [1]. Intestinal absorption has been characterized in Caco-2 monolayer permeability assays and in vivo mouse pharmacokinetic studies. Following oral administration of 36 mg/kg to mice, R-13 produces a maximum plasma concentration (C_{max}) of 129 ng/mL at a time to peak (T_{max}) of 30 minutes [1]. Oral bioavailability is approximately 10.5 percent, representing a 2.3-fold improvement over the parent 7,8-DHF (4.6 percent) [1].

5.2 Distribution

7,8-DHF released from R-13 distributes to the brain, with concentrations exceeding 8 ng/g detected at 2 hours and persisting above quantifiable levels for at least 4 hours after a single oral dose [1]. Brain-to-plasma partition coefficients for 7,8-DHF in neonatal mouse studies have been reported at approximately 8.6 to 9.9 percent [20]. The compound crosses the blood-brain barrier, supporting central nervous system pharmacological activity. Volume of distribution data for R-13 itself have not been separately reported; the distribution is dominated by the released 7,8-DHF metabolite.

5.3 Metabolism

R-13 undergoes hepatic hydrolysis as its primary metabolic transformation. The bis-carbamate is cleaved sequentially by hepatic esterases to release a monophenol intermediate (T1) and then free 7,8-DHF [1]. The liberated 7,8-DHF is subsequently metabolized by the same phase II pathways that limit its oral bioavailability when administered directly: glucuronidation at the 7- and 8-hydroxyl positions, sulfation, and catechol-O-methyltransferase-mediated O-methylation to produce 8-hydroxy-7-methoxyflavone and 7-hydroxy-8-methoxyflavone [3, 7]. Both O-methylated metab-

olites retain partial TrkB agonist activity in vitro but at reduced potency compared to the parent 7,8-DHF [3]. The specific cytochrome P450 isoforms involved in oxidative metabolism of R-13 or 7,8-DHF have not been formally characterized in a published CYP reaction phenotyping study. No striking CYP polymorphic phenotype has been identified, contrasting with compounds such as tropisetron that exhibit CYP2D6-dependent pharmacokinetic variability.

5.4 Elimination

The plasma elimination half-life of R-13 in mice is approximately 220 minutes (3.66 hours) after oral administration, compared to 134 minutes (2.23 hours) for the parent 7,8-DHF [1]. The area under the plasma concentration-time curve (AUC 0 to infinity) for R-13 at 36 mg/kg oral is 11,880 min ng/mL [1]. Elimination is predominantly metabolic (hepatic hydrolysis and phase II conjugation) rather than renal. Biliary and renal excretion of conjugated metabolites has not been quantified in published reports.

5.5 Routes of administration

The clinical route of administration is oral. In preclinical studies, R-13 has been administered exclusively by the oral route, dissolved in 5 percent dimethyl sulfoxide and 0.5 percent methylcellulose vehicle [1, 11, 12]. The oral formulation developed for clinical evaluation by Braegen Pharmaceutical is a tablet [15]. Intravenous and intraperitoneal routes have been used for the parent 7,8-DHF but not for R-13, consistent with the compound's design as an oral prodrug.

5.6 Dose translation

Preclinical doses in mice range from 7.25 to 43.6 mg/kg/day, corresponding to approximately 5, 15, and 30 mg/kg equivalents of 7,8-DHF respectively [1]. Allometric dose translation from mouse to human using the standard body surface area conversion factor (mouse dose divided by 12.3) yields approximate human-equivalent doses of 0.6 to 3.5 mg/kg, or 42 to 245 mg for a 70-kg adult. Clinical doses evaluated in the Phase 1 trial have not been published in peer-reviewed form as of this revision.

6. Clinical Evidence Base

6.1 Phase 1 clinical trial in Alzheimer's disease

Braegen Pharmaceutical completed a Phase 1 clinical trial of BrAD-R13 (R-13 formulated as an oral tablet) for Alzheimer's disease [15]. The trial was described as the first clinical evaluation of a medicine in this mechanistic class (TrkB agonist prodrug) to reach the clinical stage [15]. BrAD-R13 was reported to be well tolerated, with early results suggesting potential for brain cell protection and improvement of memory and learning [15]. Detailed clinical pharmacokinetic parameters, adverse-event rates, dose-finding data, and biomarker results from the Phase 1 trial have not been published in a peer-reviewed journal as of the most recent monograph revision. Phase 2 trials are planned to evaluate efficacy and long-term safety.

6.2 No other clinical indications

R-13 has not been evaluated in human clinical trials for any indication other than Alzheimer's disease. Preclinical development for amyotrophic lateral sclerosis, Parkinson's disease, depressive disorders, schizophrenia, and peripheral nerve injury is ongoing, but none of these indications has reached the clinical stage [2, 11, 12].

6.3 Preclinical evidence supporting clinical translation

The preclinical evidence base for R-13 is substantial and spans multiple disease models. The clinical translation rationale rests on: (a) dose-dependent efficacy in the 5XFAD Alzheimer's disease model at doses that translate to plausible human oral doses [1]; (b) absence of detectable toxicity at the highest tested dose over 12 weeks of chronic administration [1]; (c) demonstration of species-appropriate pro-drug activation in human liver microsomes [1]; (d) oral tablet formulation feasibility for long-term administration in elderly patients [15]; and (e) a mechanism (BDNF/TrkB pathway restoration) that addresses a validated pathological axis in Alzheimer's disease rather than a single downstream biomarker [9, 10].

7. Preclinical Pharmacology

7.1 Alzheimer's disease: 5XFAD transgenic mouse model

The principal preclinical demonstration was conducted by Chen et al. (2018) in the 5XFAD transgenic mouse model, which expresses five familial Alzheimer's disease mutations (Swedish, Florida, London in APP; M146L and L286V in presenilin 1) and develops aggressive amyloid pathology beginning at approximately 2 months of age [1].

R-13 was administered orally at three doses (7.25, 21.8, and 43.6 mg/kg/day) for 3 months beginning at 2 months of age. The 21.8 mg/kg dose (approximately 15 mg/kg 7,8-DHF equivalent) was identified as the optimal dose on the basis of the overall therapeutic profile [1]. Key findings at this dose included: significant reduction in amyloid-beta 40 concentration in brain homogenates; dose-dependent reduction in senile plaque deposition quantified by thioflavin S and anti-amyloid-beta immunostaining; restoration of dendritic spine density confirmed by Golgi staining; enhanced long-term potentiation in hippocampal slices; reduced interleukin-1-beta, interleukin-6, and tumor necrosis factor alpha levels; inhibition of AEP enzymatic activity; and reversal of spatial memory deficits in the Morris water maze (reduced latency to platform, increased time in target quadrant) [1].

At the molecular level, R-13 treatment potently activated TrkB phosphorylation and downstream Akt and ERK/MAPK signaling in brain tissue, confirming the in vivo pharmacodynamic mechanism. AEP phosphorylation at threonine 322 was increased dose-dependently, consistent with Akt-mediated inactivation [1, 9].

7.2 Mitochondrial neuroprotection in 5XFAD mice

A follow-up study by Chen et al. (2021) characterized the mitochondrial mechanism of R-13 neuroprotection in 5XFAD mice, reporting that R-13 treatment restored expression of oxidative phosphorylation complex proteins, improved mitochondrial membrane potential, and reduced reactive oxygen species generation in hippocampal neurons, effects mediated through TrkB/Akt signaling to the PGC-1-alpha transcriptional coactivator [21].

7.3 Amyotrophic lateral sclerosis: SOD1-G93A transgenic mouse model

Li et al. (2021) administered R-13 orally to SOD1-G93A transgenic mice (the standard preclinical model of familial ALS expressing human superoxide dismutase 1 with the G93A point mutation) beginning at postnatal day 40 and continuing for 90 days [11]. Three dose groups were evaluated: 3.6 mg/kg (n=9), 21.8 mg/kg (n=10), and 43.6 mg/kg (n=12), compared to vehicle-treated transgenic controls (n=9) and non-transgenic wild-type littermates (n=10).

R-13 significantly attenuated abnormal motor performance on rotarod and climbing-pole tests, though grip strength and hanging endurance were not significantly improved, suggesting selective protection of complex coordinated motor functions [11]. Histological analysis demonstrated that R-13 significantly preserved spinal motor neuron counts, reduced gastrocnemius muscle cross-sectional area loss, and attenuated microglial (Iba1-positive) and astrocytic (GFAP-positive) proliferation in the spinal cord [11].

The mitochondrial mechanism was characterized in detail: R-13 activated the AMPK/PGC-1-alpha/Nrf1/Tfam cascade, increased Fis1 (a mitochondrial fission protein), upregulated oxidative phosphorylation complex proteins (Cox5a, SDHB, ATP5a), increased ATP levels in spinal cord and medulla oblongata, and decreased malondialdehyde (a lipid peroxidation marker indicating oxidative stress) [11]. In a parallel *C. elegans* model expressing G93A SOD1, treatment with 7,8-DHF significantly increased median survival from 16.1 to 19.6 days [11].

7.4 Peripheral nerve regeneration

English et al. (2022) evaluated oral R-13 in a mouse model of sciatic nerve transection and end-to-end anastomosis repair [12]. Mice received R-13 at 21.8 mg/kg daily for 2 weeks (behavioral studies) or 43.6 mg/kg (biochemical studies) beginning immediately after surgery.

R-13 produced rapid TrkB phosphorylation (within 15 minutes, persisting for 8 hours) and sustained ERK/MAPK activation [12]. At 4 weeks post-injury, R-13-treated mice showed significantly greater numbers of labeled motor neurons in gastrocnemius and tibialis anterior muscles, approaching counts observed in intact uninjured animals. Sensory neuron regeneration was similarly enhanced across all size classes [12]. Electromyographic assessment revealed that direct muscle re-

sponse (M-wave) amplitudes were 2- to 3-fold greater in R-13-treated animals compared to vehicle controls or oral 7,8-DHF-treated animals [12]. The superiority of R-13 over the parent 7,8-DHF (which showed no significant advantage over vehicle at the same effective dose) confirmed the prodrug strategy as necessary for efficacy via the oral route.

7.5 Bone metabolism

Xiong et al. (2022) characterized R-13 in an ovariectomy-induced osteoporosis model in mice [13]. R-13 treatment activated TrkB/Akt signaling in bone marrow stromal cells, phosphorylated and inactivated AEP, and upregulated osteoprotegerin (OPG) through CREB-dependent transcription. Concurrently, the C/EBP-beta/AEP pathway was repressed, inhibiting RANKL-induced osteoclast differentiation. R-13 elevated OPG without altering RANKL levels and blocked trabecular bone loss with efficacy equivalent to anti-RANKL antibody treatment [13]. This work was published in Nature Communications and represents an extension of the R-13 pharmacology to musculoskeletal biology through the same TrkB/Akt/AEP axis characterized in the neurodegenerative disease models.

7.6 Depression and neuropsychiatric models

The parent compound 7,8-DHF has been extensively characterized in rodent models of depression (forced swim test, tail suspension test, chronic unpredictable stress), post-traumatic stress disorder, and schizophrenia, with consistent antidepressant-like and anxiolytic-like effects mediated through TrkB signaling [3, 22]. R-13 is under preclinical evaluation for depressive disorders and schizophrenia, though published studies using R-13 (rather than 7,8-DHF) in these models are limited as of this revision [2].

8. Sourcing and Quality Verification

Research-grade R-13 is supplied by multiple chemical and pharmaceutical research suppliers. Identified sources include Cayman Chemical (catalog number 42074, CAS 1609067-49-3), Axon Medchem (Axon 3775), and Sundia MediTech (Shanghai, China; the original synthesis contractor for the Chen/Ye laboratory) [5, 6]. Purity is typically reported as greater than 95 percent by HPLC.

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

- High-performance liquid chromatography area-percent purity, ideally greater than 95 percent (greater than 98 percent preferred for dose-response precision)
- Mass spectrometry confirmation of molecular weight (expected $[M+H]^+$ at m/z 369.108 for the protonated molecular ion)
- Nuclear magnetic resonance spectroscopy (proton and carbon-13) to confirm the bis-carbamate substitution pattern and the absence of free catechol (which would indicate premature hydrolysis)
- Differential scanning calorimetry or melting point determination for batch-to-batch consistency
- Karl Fischer or loss-on-drying analysis for residual water content
- Residual solvent analysis for common synthesis solvents (dichloromethane, ethyl acetate, dimethyl sulfoxide, methanol)

The research-grade R-13 market is smaller than that for well-established pharmaceutical compounds, and lot sizes are typically in the milligram to low-gram range. Pricing is moderate to high, in the range of \$100 to \$500 per 50 mg depending on supplier and quantity. Investigators conducting chronic dosing studies in rodents should secure sufficient material in a single lot to complete the study, as lot-to-lot variability in purity and in carbamate hydrolysis kinetics may introduce dose-response noise.

Clinical-grade BrAD-R13 (tablet formulation) is manufactured by Braegen Pharmaceutical and is not commercially available; access is through clinical trial enrollment [15].

9. Reconstitution and Handling

R-13 is supplied as a solid (off-white to pale yellow powder). The compound is soluble in dimethyl sulfoxide (DMSO) and is typically reconstituted for in vivo research use in a vehicle of 5 percent DMSO in 0.5 percent methylcellulose in water [1, 11, 12]. This vehicle produces a suspension suitable for oral gavage in mice and rats.

For in vitro studies, stock solutions are prepared in pure DMSO at concentrations of 10 to 50 mM (corresponding to approximately 3.7 to 18.4 mg/mL). Final DMSO concentrations in cell culture media should not exceed 0.1 to 0.5 percent to avoid solvent toxicity artifacts.

R-13 is less water-soluble than the parent 7,8-DHF and requires organic cosolvent for dissolution. Aqueous stock solutions are not recommended and should not be stored for more than one day owing to the risk of hydrolysis in solution [1]. The carbamate linkage is stable at acidic pH (pH 1.2) but susceptible to base-catalyzed hydrolysis; solutions should be maintained at neutral or mildly acidic pH.

Storage of the solid compound at minus 20 degrees Celsius in a desiccated container, protected from light, is recommended by commercial suppliers. The solid is stable under these conditions for at least 2 years. Solutions in DMSO may be stored at minus 20 degrees Celsius for up to 6 months in single-use aliquots to avoid freeze-thaw cycles.

Investigators should monitor hydrolysis during extended in vivo dosing studies by periodic HPLC analysis of the dosing vehicle to confirm that R-13 has not degraded to 7,8-DHF prior to administration.

10. Stack Interaction Considerations

R-13 interacts with other pharmacological agents through its TrkB/BDNF-pathway mechanism, its hepatic ester hydrolysis, and the phase II metabolism of the released 7,8-DHF. Investigators conducting research with R-13 in combination with other compounds should consider:

Esterase inhibitors: Compounds that inhibit hepatic carboxylesterases (organophosphates, bis-p-nitrophenyl phosphate) may reduce the conversion of R-13 to 7,8-DHF and diminish efficacy. Concurrent administration of esterase inhibitors is not recommended in R-13 research protocols.

Catechol-O-methyltransferase (COMT) inhibitors: Entacapone, tolcapone, and opicapone inhibit the O-methylation of catechol-containing compounds and may increase systemic 7,8-DHF exposure when co-administered with R-13. This interaction is theoretically synergistic and may be of research interest in Parkinson's disease models where COMT inhibitors are already part of the therapeutic regimen.

Phase II enzyme inducers: Compounds that induce UDP-glucuronosyltransferases or sulfotransferases (rifampin, phenobarbital, phenytoin, carbamazepine) may accelerate 7,8-DHF conjugation and reduce effective exposure.

Other TrkB agonists or BDNF-pathway modulators: Co-administration with other TrkB ligands (LM22A-4, deoxygedunin, amitriptyline at TrkB-activating concentrations) produces additive effects on TrkB signaling and may exceed the physiological range of downstream pathway activation. Combination studies should include dose-response characterization to identify synergistic or antagonistic interactions.

Cholinesterase inhibitors: Concurrent donepezil, rivastigmine, or galantamine (standard-of-care Alzheimer's disease medications) with R-13 is a likely clinical scenario. The parent 7,8-DHF has been co-administered with donepezil in preclinical models without characterized antagonism; the TrkB mechanism is independent of cholinergic transmission and the combination is theoretically complementary.

Anti-amyloid antibodies: Concurrent lecanemab, donanemab, or aducanumab with R-13 represents a plausible combination strategy targeting both amyloid clearance (antibody) and amyloid production/tau pathology (R-13/AEP inhibition). No published interaction data exist.

Antioxidants: The flavone core of 7,8-DHF has intrinsic radical-scavenging activity. Co-administration with other polyphenolic antioxidants (resveratrol, curcumin, epigallocatechin gallate) may produce additive antioxidant effects but also competitive interactions for phase II conjugation enzymes, potentially altering the pharmacokinetic profile of 7,8-DHF.

Serotonergic agents: No direct interaction with serotonergic pathways is predicted from the TrkB mechanism. However, BDNF signaling modulates serotonin receptor expression and serotonergic neurotransmission in the hippocampus and prefrontal cortex; investigators combining R-13 with SSRIs or SNRIs should monitor for augmented antidepressant-like effects in preclinical behavioral assays.

11. Adverse Events and Safety

11.1 Preclinical toxicity

The most comprehensive toxicity assessment is the Chen et al. (2018) chronic study in wild-type C57BL/6 mice receiving R-13 at 43.6 mg/kg/day (the highest tested dose, approximately 30 mg/kg 7,8-DHF equivalent) by oral gavage for 12 consecutive weeks [1]. No detectable organ toxicity was reported across liver, kidney, heart, lung, spleen, brain, or gastrointestinal tract by histopathological examination. Body weight trajectories did not differ from vehicle controls. Serum chemistry panels (alanine aminotransferase, aspartate aminotransferase, blood urea nitrogen, creatinine) were within normal ranges [1].

The parent compound 7,8-DHF has similarly demonstrated no detectable adverse effects in chronic dosing studies in mice and rats across multiple laboratories over approximately 15 years of preclinical investigation [3, 4].

11.2 Theoretical safety considerations

TrkB-mediated proliferative risk. TrkB is expressed on certain tumors, including neuroblastoma and medulloblastoma. Chronic TrkB agonism could theoretically promote tumor cell survival or proliferation in individuals with occult TrkB-expressing malignancies. This concern is theoretical; no tumor promotion has been observed in preclinical chronic dosing studies at studied doses and durations [1, 3].

Cardiovascular effects. TrkB is expressed on cardiac myocytes and endothelial cells. BDNF/TrkB signaling has been implicated in cardiac angiogenesis and in the modulation of cardiac sympathetic innervation. Chronic TrkB agonism has not produced detectable cardiovascular adverse effects in preclinical studies, but dedicated cardiovascular safety pharmacology (hERG channel assay, telemetered dog or non-human primate cardiovascular study) has not been published for R-13.

Seizure risk. BDNF/TrkB signaling is implicated in epileptogenesis; chronic TrkB activation theoretically carries a risk of lowering seizure threshold, particularly in models of temporal lobe epilepsy. However, 7,8-DHF at therapeutic doses has

demonstrated anticonvulsant rather than proconvulsant effects in preclinical seizure models, and R-13 has not been associated with seizure activity [3, 23].

11.3 Clinical safety

The Phase 1 trial of BrAD-R13 was reported to demonstrate tolerability, but detailed adverse-event data (rates, severity grades, dose-limiting toxicities, laboratory abnormalities) have not been published in peer-reviewed form [15]. The oral tablet formulation was described as practical for long-term use in elderly patients, suggesting an acceptable gastrointestinal tolerability profile in the Phase 1 population.

11.4 Pregnancy and developmental toxicity

No published reproductive or developmental toxicity studies of R-13 are available. The parent 7,8-DHF has not been formally evaluated in standard reproductive toxicity paradigms. Use in pregnancy or lactation is not recommended absent safety data.

12. Comparable Compounds Assessment

This section presents a structured comparative assessment of five TrkB agonist and BDNF-mimetic candidates against R-13, evaluated against five competency standards: novelty, effect size, promising potential (current clinical state and trajectory), side-effect profile, and overall validation. The five comparators were selected as the principal TrkB-targeted or BDNF-pathway-modulating compounds with substantive preclinical or clinical pharmacology programs.

12.1 7,8-Dihydroxyflavone (7,8-DHF; tropoflavin)

Mechanism: Direct small-molecule TrkB agonist. 7,8-DHF binds the TrkB extracellular domain and activates receptor dimerization and autophosphorylation [4]. The compound is the parent molecule from which R-13 is derived.

Development trajectory: 7,8-DHF was the first small-molecule TrkB agonist identified (Jang et al. 2010, PNAS) and has been evaluated in hundreds of preclinical studies across neurodegeneration, stroke, depression, traumatic brain injury, hearing loss, and metabolic disease over approximately 15 years [3, 4]. The compound has not been advanced to human clinical trials owing to poor oral bioavailability (approximately 4.6 percent) and rapid hepatic conjugation [1, 7]. It is widely sold as a dietary supplement in capsule form (typically 25 mg), though no human dosing studies have established safety, efficacy, or pharmacokinetics.

Novelty: Moderate. The compound established the TrkB small-molecule agonist field and remains the reference compound. The mechanism was novel at the time of discovery (2010) but is now well established.

Effect size: Robust in preclinical models across many indications. Effect sizes in Alzheimer's disease models (5XFAD, APP/PS1) are substantial for amyloid reduction and cognitive rescue [3, 24]. No human data.

Promising potential: Low as a standalone clinical candidate owing to poor oral bioavailability. High as a research tool and as the pharmacologically active metabolite of prodrug candidates (R-13, CF3CN).

Side-effect profile: Favorable. No detectable toxicity in chronic preclinical studies [3, 4]. No human safety data.

Overall validation: High as a research tool; low as a clinical candidate. The most extensively validated TrkB agonist in preclinical pharmacology.

Comparison to R-13: R-13 was specifically designed to overcome the pharmacokinetic limitations of 7,8-DHF while preserving its receptor pharmacology. R-13 demonstrated oral bioavailability 2.3-fold greater than 7,8-DHF, a longer elimination half-life (220 versus 134 minutes), and superior efficacy in the peripheral nerve regeneration model where oral 7,8-DHF failed to outperform vehicle [1, 12]. R-13 has advanced to clinical evaluation whereas 7,8-DHF has not.

12.2 CF3CN (optimized TrkB agonist)

Mechanism: Synthetic derivative of 7,8-DHF that interacts with the TrkB LRM/CC2 domain with improved druglike properties [16]. CF3CN is not a prodrug of 7,8-DHF but a direct-acting TrkB agonist with structural modifications to the flavone core.

Development trajectory: Chen et al. (2021) characterized CF3CN as a second-generation optimized TrkB agonist with improved oral bioavailability and half-life compared to 7,8-DHF [16]. In 5XFAD mice, oral CF3CN enhanced BDNF levels, phosphorylated AEP at T322, antagonized delta-secretase activity, attenuated amyloid and tau pathology, and improved cognitive function. Chronic treatment demonstrated no detectable toxicity [16].

Novelty: High within the TrkB agonist class. The structural optimization represents a distinct approach from the prodrug strategy of R-13.

Effect size: Comparable to or modestly greater than R-13 in the 5XFAD model based on published data [16].

Promising potential: Moderate. The compound is preclinical and has not advanced to clinical trials. R-13 was selected as the lead clinical candidate by Braegen Pharmaceutical, suggesting that R-13 was preferred on the balance of properties despite the optimization of CF3CN.

Side-effect profile: No detectable toxicity in chronic preclinical studies [16]. No human safety data.

Overall validation: Low to moderate. Published evidence is limited to a single primary publication.

Comparison to R-13: CF3CN and R-13 are parallel optimization strategies from the same laboratory. R-13 is a prodrug that releases 7,8-DHF; CF3CN is a direct-acting modified agonist. R-13 has the advantage of clinical advancement; CF3CN has the theoretical advantage of not requiring prodrug activation but has not been advanced to the clinic.

12.3 LM22A-4

Mechanism: Non-peptide small molecule reported as a partial TrkB agonist and BDNF mimetic. LM22A-4 was identified through a computational screen of BDNF loop domain-mimicking structures [25]. It is 98 percent smaller than BDNF (molecular weight approximately 302 g/mol versus approximately 27 kDa) and was reported to activate TrkB and promote neuronal survival.

Development trajectory: LM22A-4 has been characterized in preclinical models of traumatic brain injury, stroke, Huntington's disease, Rett syndrome, multiple sclerosis (myelin repair), and optic neuropathy [25, 26, 27]. It has not been advanced to human clinical trials.

Novelty: Moderate. The computational design approach was novel at the time of discovery, but the compound has been available as a research tool for over a decade.

Effect size: Variable. Neuroprotective effects in traumatic brain injury and Rett syndrome models are reproducible. Effect sizes in Alzheimer's disease models are less well characterized than those of R-13 or 7,8-DHF.

Promising potential: Low to moderate. No clinical development. The compound is principally a research tool.

Side-effect profile: No reported toxicity in preclinical studies. No human safety data.

Overall validation: Moderate, but the mechanistic basis has been questioned. Bhatt et al. (2017) reported that LM22A-4 may not directly activate TrkB but instead produce delayed TrkB transactivation through a GPCR-mediated mechanism [19].

This mechanistic ambiguity reduces confidence in the specificity of the compound as a TrkB agonist.

Comparison to R-13: R-13 has the advantages of a clearer mechanistic basis (pro-drug releasing the well-characterized TrkB agonist 7,8-DHF), more extensive pre-clinical characterization in Alzheimer's disease, and clinical advancement. LM22A-4 has the advantage of direct (non-prodrug) mechanism and does not require hepatic activation, which could be advantageous in models of hepatic impairment.

12.4 Deoxygedunin

Mechanism: Natural product (limonoid terpenoid) isolated from the Indian neem tree (*Azadirachta indica*) and reported to activate TrkB in a BDNF-independent manner [28]. Deoxygedunin binds TrkB at a site distinct from the BDNF binding site and provokes receptor autophosphorylation and downstream Akt and ERK activation.

Development trajectory: Deoxygedunin has been characterized primarily in vitro and in rodent models of neurodegeneration and hearing loss. Jang et al. (2010) and subsequent reports demonstrated neuroprotective effects in kainic acid excitotoxicity and ototoxicity models [28]. The compound has not been advanced beyond early-stage preclinical evaluation.

Novelty: Moderate. The natural product origin and the BDNF-independent TrkB activation mechanism are distinctive.

Effect size: Moderate in excitotoxicity and hearing loss models. Limited Alzheimer's disease-specific data.

Promising potential: Low. No clinical development program. Limited recent pre-clinical publications.

Side-effect profile: Limited safety characterization. The limonoid scaffold carries potential for hepatotoxicity at high doses, though this has not been formally characterized for deoxygedunin.

Overall validation: Low. The compound is a research tool with limited validation relative to 7,8-DHF, R-13, or LM22A-4.

Comparison to R-13: R-13 has substantially greater preclinical validation, a clearer translational pathway, and clinical advancement. Deoxygedunin is principally of interest as a mechanistic probe for BDNF-independent TrkB activation.

12.5 PHA-543613

Mechanism: Highly selective alpha-7 nicotinic acetylcholine receptor (nAChR) agonist. PHA-543613 does not directly target TrkB but is included in this comparator panel because alpha-7 nAChR activation modulates BDNF expression and TrkB signaling through downstream transcriptional mechanisms [29, 30]. PHA-543613 is a Pfizer research compound with full agonist activity at alpha-7 nAChR, good brain penetration, and oral bioavailability in rodents.

Development trajectory: PHA-543613 has been characterized in rodent cognitive models, including scopolamine-induced amnesia, presenilin double-knockout Alzheimer models, and aged-rat working memory paradigms [29, 30]. Synergistic cognitive effects with memantine in aged rats have been reported [30]. The compound has not been advanced to human clinical trials.

Novelty: Low to moderate within the alpha-7 nAChR agonist class; the connection to BDNF/TrkB pathway is indirect and mechanistically distinct from direct TrkB agonism.

Effect size: Substantial in preclinical cognitive models, particularly in combination with memantine [30]. However, direct comparison with R-13 is confounded by the different receptor targets and downstream mechanisms.

Promising potential: Limited as a clinical candidate. Substantial as a research tool for understanding the intersection of cholinergic and neurotrophic signaling.

Side-effect profile: Preclinical only. No human safety data. Rodent safety has been adequate.

Overall validation: Low. Principally a preclinical research tool without clinical validation.

Comparison to R-13: R-13 targets TrkB directly through its active metabolite 7,8-DHF, producing immediate receptor activation without dependence on transcriptional upregulation of BDNF. PHA-543613 acts upstream through alpha-7 nAChR to modulate BDNF expression, an indirect and slower-onset mechanism. R-13 has clin-

ical advancement; PHA-543613 does not. The two compounds could be complementary in combination studies aimed at both direct TrkB activation (R-13) and enhanced endogenous BDNF production (PHA-543613).

12.6 Comparator summary

The five compounds occupy distinct positions in the TrkB/BDNF signaling pharmacology space. **7,8-DHF** is the most extensively validated preclinical TrkB agonist and the active metabolite of R-13, limited only by poor oral bioavailability. **CF3CN** is a second-generation direct-acting TrkB agonist with improved pharmacokinetics but no clinical advancement. **LM22A-4** is a computationally designed BDNF mimetic with mechanistic ambiguity. **Deoxygedunin** is a natural product TrkB agonist with limited validation. **PHA-543613** represents an indirect BDNF-modulatory approach through alpha-7 nAChR.

R-13 occupies a unique position in this landscape: it is the only compound with an active clinical development program, the only prodrug strategy that has demonstrated superiority over its parent compound in a head-to-head in vivo comparison (peripheral nerve regeneration), and the compound with the most comprehensive preclinical dossier spanning Alzheimer's disease, ALS, bone metabolism, and nerve regeneration. Its principal limitation is the early clinical stage: Phase 1 data remain unpublished in peer-reviewed form, and Phase 2 efficacy has not been demonstrated. The mechanistic foundation (TrkB/Akt/AEP axis) is well validated across multiple laboratories and disease models, and the prodrug design addresses the principal translational barrier that has prevented the parent 7,8-DHF from reaching the clinic despite 15 years of preclinical evidence.

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tain analytical confirmation of identity and purity on every research-grade lot. The parent compound 7,8-dihydroxyflavone is sold as a dietary supplement in some markets but has not been evaluated in controlled human clinical trials for safety or efficacy.