

RESEARCH MONOGRAPH . COMPOUND PROFILE

BAM15

A synthetic oxadiazolopyrazine-scaffold mitochondrial protonophore distinguished from classical uncouplers by selective dissipation of the inner mitochondrial membrane proton gradient without depolarization of the plasma membrane, conferring potent metabolic enhancement with markedly reduced cytotoxicity in preclinical models of obesity, insulin resistance, hepatic steatosis, sepsis, and cancer.

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ABSTRACT

BAM15 (N5,N6-bis(2-fluorophenyl)-[1,2,5]oxadiazolo[3,4-b]pyrazine-5,6-diamine) is a synthetic small-molecule mitochondrial protonophore first identified in a 2014 phenotypic screen by Kenwood et al. at the University of Virginia and Virginia Tech for compounds that uncouple mitochondrial oxidative phosphorylation without depolarizing the plasma membrane [1]. The compound dissipates the electrochemical proton gradient across the inner mitochondrial membrane, thereby uncoupling electron transport from adenosine triphosphate (ATP) synthesis and increasing substrate oxidation and energy expenditure. Unlike the classical protonophore uncouplers 2,4-dinitrophenol (DNP) and carbonylcyanide-p-trifluoromethoxyphenylhydrazone (FCCP), BAM15 selectively targets the mitochondrial membrane and does not collapse the plasma membrane potential at effective uncoupling concentrations, a property that confers a substantially wider therapeutic index and reduced cytotoxicity in cultured cells and in vivo [1, 2]. The compound is orally bioavailable in mice (67 percent oral bioavailability, C_{max} 8.2 micromolar, t_{1/2} 1.7 hours) with primary distribution to the liver, supporting hepatic metabolic applications [3]. In C57BL/6J mice fed a high-fat diet, BAM15 administered at 100 mg/kg/day by oral gavage reversed diet-induced obesity, decreased body fat mass without altering food intake or lean body mass, reduced hepatic triglycerides by approximately 75 percent, decreased inflammatory lipids, and improved whole-body insulin sensitivity as demonstrated by hyperinsulinemic-euglycemic clamp [3]. In a head-to-head comparison in female db/db mice, BAM15 and calorie restriction improved body weight and liver steatosis to levels superior to

semaglutide, niclosamide ethanolamine (NEN), and rosiglitazone, while BAM15, semaglutide, and rosiglitazone completely restored glucose tolerance [4]. These metabolic effects are mediated through sustained activation of AMP-activated protein kinase (AMPK) and peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC-1alpha), promoting fatty acid oxidation, glucose uptake, and mitochondrial biogenesis [2, 5]. Beyond metabolic disease, BAM15 has demonstrated preclinical efficacy in acute kidney injury and sepsis (reducing mortality even when administered 12 hours after cecal ligation and puncture in mice) [6], in acute myeloid leukemia (inhibiting AML cell proliferation and inducing reactive oxygen species-mediated apoptosis with selectivity over normal cells) [7], in sarcopenic obesity (preserving skeletal muscle contractility and mitochondrial respiration in aged mice) [8, 9], in atherosclerosis (suppressing western diet-induced plaque formation in ApoE-knockout mice through AMPK activation and NF-kappaB/NLRP3 inflammasome suppression) [10, 11], and in vascular smooth muscle relaxation [5]. Safety pharmacology in rodents has demonstrated no alteration of body temperature, food intake, lean body mass, or standard hematological and biochemical markers of toxicity at effective metabolic doses [3]. BAM15 has not entered human clinical trials as of the most recent monograph revision. The compound is supplied as a research-grade material by multiple chemical suppliers at greater than 98 percent purity and is not approved by any regulatory authority for therapeutic use. This monograph documents the chemistry, synthesis, discovery history, molecular pharmacology, pharmacokinetics, preclinical evidence base across metabolic, inflammatory, oncologic, and aging indications, sourcing and quality verification, reconstitution and handling, stack-interaction considerations, adverse-event profile, and a

structured comparative assessment against five alternative mitochondrial uncouplers.

1. Compound Identification

1.1 Names, synonyms, and registry identifiers

Preferred name: BAM15.

Systematic chemical name (International Union of Pure and Applied Chemistry): N5,N6-bis(2-fluorophenyl)-[1,2,5]oxadiazolo[3,4-b]pyrazine-5,6-diamine.

Chemical Abstracts Service registry number: 210302-17-3.

PubChem compound identifier: CID 565708.

Synonyms and alternative designations: BAM 15, BAM-15. The compound is referred to by the BAM15 designation in essentially all published literature; no international nonproprietary name or marketed trade name has been assigned, as the compound has not entered clinical development.

1.2 Structural class and chemistry

BAM15 belongs to the [1,2,5]oxadiazolo[3,4-b]pyrazine class of heterocyclic compounds, specifically the 5,6-diaminoxadiazolopyrazine subclass bearing N-aryl substituents. The core scaffold consists of a fused bicyclic system comprising a 1,2,5-oxadiazole ring (furan) and a pyrazine ring, with two 2-fluoroanilino groups appended at the 5- and 6-positions through secondary amine linkages. The compound is a symmetric bisanilino derivative; the N-H protons of the aniline linkages are essential for protonophore activity, as they participate in proton shuttling across the lipid bilayer of the inner mitochondrial membrane [12, 13].

Molecular formula: C₁₆H₁₀F₂N₆O.

Molecular weight: 340.29 g/mol (monoisotopic exact mass: 340.0885).

Physical properties: Off-white to pale yellow crystalline powder. Low aqueous solubility consistent with the lipophilic character required for membrane partitioning and protonophore function. Soluble in dimethyl sulfoxide (DMSO) at con-

centrations of 10 to 50 mM. Sparingly soluble in ethanol. The low aqueous solubility does not limit oral bioavailability in vivo, as the compound is absorbed through the gastrointestinal epithelium with 67 percent bioavailability in mice [3].

Log P: Estimated octanol-water partition coefficient consistent with moderate lipophilicity (cLogP approximately 3.5 to 4.0), supporting membrane partitioning and mitochondrial localization.

1.3 Synthesis

The synthesis of BAM15 follows a three-step sequence from commercially available starting materials. The initial step involves the reaction of 3,4-diaminofurazan (diaminoxadiazole) with oxalic acid under acidic conditions (aqueous hydrochloric acid) to form the fused oxadiazolopyrazine-diol intermediate. The second step is chlorination of the diol intermediate with phosphorus pentachloride and phosphorus oxychloride (PCl₅/POCl₃) to afford the 5,6-dichloro-[1,2,5]oxadiazolo[3,4-b]pyrazine intermediate. The third step is nucleophilic aromatic substitution of both chlorines with 2-fluoroaniline in the presence of triethylamine in tetrahydrofuran (THF) under reflux conditions overnight, yielding BAM15 as the symmetric bis(2-fluoroanilino) product [12, 13]. The product is purified by column chromatography or recrystallization. Identity is confirmed by proton and carbon-13 nuclear magnetic resonance spectroscopy, mass spectrometry, and high-performance liquid chromatography. Research-grade material is typically supplied at greater than 95 to 98 percent purity.

Structure-activity relationship studies on the oxadiazolopyrazine scaffold have demonstrated that the N-H proton is essential for protonophore activity; N-methylation abolishes uncoupling. Electron-withdrawing groups on the aniline rings are preferred for potency. Asymmetric derivatives bearing hydroxylamine or hydrazine substituents in place of one aniline group have been identified as more potent uncouplers than BAM15 itself [13, 14]. Evolution of the scaffold to the related oxadiazolopyridine core yielded the derivative SHM115, which demonstrates improved tissue distribution to white and brown adipose tissue relative to BAM15 [15].

2. Abstract

BAM15 is a synthetic mitochondrial protonophore uncoupler that dissipates the proton gradient across the inner mitochondrial membrane without depolarizing the plasma membrane, a selectivity feature that distinguishes it from the classical uncouplers 2,4-dinitrophenol and FCCP and confers a substantially reduced cytotoxicity profile [1]. The compound was identified in 2014 through a cell-based phenotypic screen designed to discover uncouplers with a broad effective concentration range and absent plasma membrane activity [1]. In cultured cells, BAM15 stimulates a higher maximal rate of mitochondrial respiration than FCCP at equimolar concentrations, produces sustained respiratory stimulation with a longer half-life of activity, and does not induce caspase 3/7 activation (a marker of apoptotic cell death) at concentrations up to 40 micromolar, whereas both FCCP and DNP activate caspases at lower concentrations [1, 2].

The preclinical pharmacology of BAM15 spans metabolic disease, hepatic steatosis, sepsis and acute kidney injury, oncology, atherosclerosis, sarcopenia, and aging. In the metabolic disease domain, oral BAM15 at 100 mg/kg/day in C57BL/6J mice fed a high-fat diet reversed obesity, reduced hepatic fat by approximately 75 percent, decreased inflammatory lipids, and improved insulin sensitivity across liver, muscle, and adipose tissue as measured by hyperinsulinemic-euglycemic clamp studies [3]. Body temperature, food intake, lean body mass, and standard toxicology markers were unaffected [3]. A head-to-head comparison in db/db mice demonstrated that BAM15 improved body weight and hepatic steatosis to levels superior to semaglutide, while completely restoring glucose tolerance [4].

The molecular mechanism involves dissipation of the mitochondrial proton motive force, which reduces the ATP/ADP ratio and activates AMPK. AMPK activation stimulates catabolic pathways including fatty acid oxidation, glucose uptake, and autophagy while suppressing anabolic pathways. BAM15 also activates PGC-1 α , promoting mitochondrial biogenesis and oxidative capacity [2, 5]. In immune cells, BAM15 suppresses NF-kappaB nuclear translocation and NLRP3 inflammasome activation, providing anti-inflammatory activity relevant to the sepsis, atherosclerosis, and chronic disease contexts [6, 10, 11].

BAM15 has not entered human clinical trials. The compound is available exclusively as a research-grade preparation and is not approved for therapeutic use in any jurisdiction. The short plasma half-life (1.7 hours in mice) and limited aqueous solubility are acknowledged pharmacokinetic limitations that have prompted medicinal chemistry optimization programs directed at improved exposure and tissue distribution [15].

3. Discovery and Development History

3.1 Origin

BAM15 was identified in 2014 by Kenwood, Bhatt, Bhatt, and colleagues at the University of Virginia and Virginia Polytechnic Institute and State University (Virginia Tech) through a cell-based phenotypic screen [1]. The screen was designed to address a longstanding problem in mitochondrial uncoupler pharmacology: the classical protonophore uncouplers DNP and FCCP, while potent stimulators of mitochondrial respiration and substrate oxidation, produce off-target depolarization of the plasma membrane at concentrations close to their effective uncoupling concentrations, resulting in cytotoxicity, electrophysiological disruption, and a dangerously narrow therapeutic index that has precluded clinical development since the withdrawal of DNP as an obesity drug in 1938 [1, 16].

The Kenwood screen evaluated compounds from chemical libraries for the ability to increase mitochondrial oxygen consumption rate (a marker of uncoupling) without depolarizing the plasma membrane potential (measured by the voltage-sensitive fluorescent dye bis-oxonol). BAM15 emerged as a hit that satisfied both criteria across a broad concentration range. Subsequent characterization demonstrated that BAM15 was equipotent to FCCP as a mitochondrial uncoupler but produced a higher maximal respiration rate in cultured cells, was substantially less cytotoxic, and was bioactive in vivo [1].

3.2 Initial in vivo demonstration

The original Kenwood et al. (2014) publication included the first in vivo demonstration: BAM15 dose-dependently protected mice from acute renal ischemia-reperfusion injury, providing the initial evidence that the mitochondrial selectivity observed in cell culture translated to an in vivo therapeutic context [1]. This renal protection finding was subsequently extended by Kellum, Parikh, and colleagues in sepsis-associated acute kidney injury models [6].

3.3 Metabolic disease development

The metabolic disease application of BAM15 was developed principally by the Webster-Santos laboratory at Virginia Tech and the Axelrod laboratory at Pennington Biomedical Research Center (Louisiana State University). Alexopoulos, Axelrod, and Webster-Santos published the seminal metabolic disease study in 2020 in *Nature Communications*, demonstrating oral BAM15 reversal of diet-induced obesity and insulin resistance in high-fat-diet-fed C57BL/6J mice [3]. Concurrently, Axelrod, Fealy, and colleagues published a complementary study in *EMBO Molecular Medicine* establishing the AMPK-dependent metabolic mechanism and extending the glycemic findings [2]. These two publications established BAM15 as a leading preclinical mitochondrial uncoupler candidate for metabolic disease.

3.4 Expansion to additional indications

Between 2020 and 2025, the BAM15 research literature expanded substantially. Major research directions include sepsis and acute kidney injury (Parikh et al. 2023, *Journal of Clinical Investigation*) [6]; acute myeloid leukemia (Gao et al. 2022) [7]; atherosclerosis (Tai et al. 2018; Liu et al. 2025) [5, 11]; sarcopenic obesity and aging (Dantas et al. 2022; ongoing bioRxiv preprint 2025) [8, 9]; NAFLD/MASH combination therapy with thyroid hormone receptor beta agonists (Zhou et al. 2024) [17]; and medicinal chemistry optimization of the oxadiazolopyrazine scaffold (Sousa et al. 2020; Harkness et al. 2025; Axelrod et al. 2023) [12, 13, 14, 15].

3.5 Current development state

BAM15 has not entered human clinical trials as of the most recent monograph revision. No investigational new drug (IND) application or clinical trial authorization has been publicly disclosed. The compound remains a preclinical research tool, though the breadth of the evidence base, the safety pharmacology data, and the ongoing medicinal chemistry optimization suggest that clinical translation is being actively pursued. Analogs with improved pharmacokinetic properties (longer half-life, improved tissue distribution) are in preclinical evaluation [15].

4. Mechanism of Action

4.1 Protonophore uncoupling of oxidative phosphorylation

The primary molecular mechanism of BAM15 is protonophore-mediated uncoupling of mitochondrial oxidative phosphorylation. Under normal physiological conditions, the electron transport chain (complexes I through IV) pumps protons from the mitochondrial matrix across the inner mitochondrial membrane into the intermembrane space, generating an electrochemical proton gradient (proton motive force, $\Delta\psi$ approximately 150 to 180 mV). This proton gradient drives ATP synthase (complex V) to synthesize ATP from ADP and inorganic phosphate. Protonophore uncouplers bypass ATP synthase by carrying protons across the inner mitochondrial membrane through the lipid bilayer, dissipating the proton gradient and converting the energy of substrate oxidation to heat rather than ATP [1, 16].

BAM15 functions as a proton shuttle. The compound partitions into the inner mitochondrial membrane, acquires a proton on the intermembrane space face (the N-H protons of the aniline linkages serve as the protonatable sites), diffuses across the membrane in the protonated form, releases the proton on the matrix face, and returns to the intermembrane space as the deprotonated species. This cycling dissipates the proton gradient in proportion to the concentration of BAM15 in the membrane [1, 12].

4.2 Mitochondrial selectivity

The defining pharmacological feature of BAM15 is its selectivity for the mitochondrial membrane over the plasma membrane. Classical protonophores (DNP, FCCP, CCCP) are relatively non-selective: they partition into any lipid bilayer and dissipate proton gradients across both the inner mitochondrial membrane and the plasma membrane. Plasma membrane depolarization produces cytotoxicity through disruption of ion homeostasis, calcium dysregulation, and activation of cell death pathways. BAM15 does not depolarize the plasma membrane at concentrations that produce maximal mitochondrial uncoupling [1]. The molecular basis of this selectivity is not fully elucidated but is attributed to preferential partitioning

into the mitochondrial membrane (which has a distinct lipid composition enriched in cardiolipin) and to the physicochemical properties of the oxadiazolopyrazine scaffold [1, 2].

In functional terms, BAM15 was well tolerated at concentrations up to 8 micromolar in cultured cells, whereas FCCP began to inhibit oxygen consumption rate at concentrations between 1 and 2 micromolar, reflecting the narrower effective range of the non-selective uncoupler [1]. BAM15 did not induce caspase 3/7 activity (a marker of apoptotic cell death) at concentrations up to 40 micromolar, while both DNP and FCCP induced caspase activity at all tested doses [1, 2].

4.3 AMPK activation

Mitochondrial uncoupling by BAM15 reduces the intracellular ATP/ADP ratio, which is the canonical stimulus for activation of AMP-activated protein kinase (AMPK). AMPK is a heterotrimeric serine/threonine kinase that functions as a cellular energy sensor. Upon activation by increased AMP/ADP relative to ATP, AMPK phosphorylates downstream substrates that stimulate catabolic energy-producing pathways (fatty acid oxidation via phosphorylation of acetyl-CoA carboxylase, glucose uptake via GLUT4 translocation, autophagy) and suppress anabolic energy-consuming pathways (lipogenesis, protein synthesis, gluconeogenesis) [2, 5].

BAM15 activates AMPK more potently than the canonical AMPK activators metformin and AICAR, and more potently than the alternative mitochondrial uncouplers CCCP and niclosamide, as demonstrated in vascular smooth muscle cells [5]. The AMPK activation is sustained during chronic BAM15 treatment and is the principal mediator of the metabolic improvements observed in the high-fat-diet mouse models [2, 3].

4.4 PGC-1alpha and mitochondrial biogenesis

Downstream of AMPK, BAM15 activates PGC-1alpha (peroxisome proliferator-activated receptor gamma coactivator 1-alpha), the master transcriptional coactivator of mitochondrial biogenesis [2]. PGC-1alpha activation upregulates nuclear-encoded mitochondrial genes, promotes mitochondrial DNA replication and transcription, and increases mitochondrial mass and oxidative capacity. This biogene-

sis response is a compensatory adaptation to the energy deficit produced by uncoupling and contributes to the sustained increase in metabolic rate observed with chronic BAM15 treatment.

4.5 Reactive oxygen species modulation

Mild mitochondrial uncoupling reduces mitochondrial superoxide production by accelerating electron flow through the electron transport chain and reducing the dwell time of electrons at complex I and complex III, the principal sites of superoxide generation [1, 3]. BAM15 therefore functions as an indirect antioxidant through its uncoupling mechanism. In the liver, this antioxidant effect contributes to the reduction of lipid peroxidation, inflammatory lipid species, and oxidative stress markers observed in high-fat-diet models [3].

Paradoxically, in cancer cells (particularly acute myeloid leukemia cells), BAM15 increases reactive oxygen species (ROS) through a mechanism that may involve severe ATP depletion and mitochondrial dysfunction in cells with high basal oxidative phosphorylation dependence. This ROS increase triggers cytochrome c release and caspase-mediated apoptosis in AML cells while sparing normal cells that have lower mitochondrial dependence [7].

4.6 Anti-inflammatory signaling

BAM15 suppresses the nuclear translocation of NF-kappaB (nuclear factor kappa-light-chain-enhancer of activated B cells) in macrophages and other immune cells, thereby inhibiting the transcription of pro-inflammatory cytokines including interleukin-6 (IL-6), interleukin-1 beta (IL-1beta), and tumor necrosis factor alpha (TNF-alpha) [6, 10, 11]. BAM15 also inhibits NLRP3 inflammasome activation, reducing caspase-1-mediated processing and secretion of IL-1beta [11]. This anti-inflammatory mechanism is relevant to the sepsis, atherosclerosis, and NAFLD/MASH applications and is mechanistically linked to AMPK activation, which is a known negative regulator of NF-kappaB signaling.

5. Pharmacokinetics and Routes of Administration

5.1 Absorption

BAM15 is orally bioavailable. In C57BL/6J mice, oral gavage of BAM15 at 50 mg/kg produced an oral bioavailability of 67 percent based on dose-normalized area under the concentration-time curve (AUC) relative to intraperitoneal administration [3]. Absorption is rapid, with maximum plasma concentrations achieved within 0.5 to 1 hour after oral dosing. The maximum plasma concentration (C_{max}) was 8.2 micromolar after a single oral dose, consistent with the low-micromolar concentrations at which BAM15 produces maximal mitochondrial uncoupling in cultured cells [3].

5.2 Distribution

BAM15 tissue distribution analysis revealed primary distribution to the liver, with gradual clearance from tissues over approximately 4 hours [3]. The hepatic concentration achieved after oral dosing supports the pronounced effects on hepatic triglyceride reduction and fatty acid oxidation observed in the metabolic disease studies. The low aqueous solubility and moderate lipophilicity of BAM15 are properties that enable membrane penetration and mitochondrial localization but may limit distribution to extrahepatic tissues at standard oral doses.

Distribution to skeletal muscle is sufficient to produce functional effects on mitochondrial respiration and contractile function in aged mice, though tissue concentrations in muscle have not been reported with the same precision as hepatic concentrations [8, 9]. Distribution to adipose tissue is modest with the parent compound; the structurally related analog SHM115 was developed specifically to improve white and brown adipose tissue distribution [15].

5.3 Metabolism

The metabolic fate of BAM15 in vivo has not been comprehensively characterized in published form. The short plasma half-life (1.7 hours) suggests rapid hepatic metabolism, likely through phase I oxidative pathways (cytochrome P450-mediated) followed by phase II conjugation. The specific CYP isoforms responsible for BAM15 metabolism have not been identified in published reports. The absence of a characterized metabolic pathway is a gap in the preclinical pharmacology that would need to be addressed prior to clinical development.

5.4 Elimination

Plasma elimination is rapid, with a half-life of approximately 1.7 hours in mice after oral administration [3]. The short half-life is acknowledged as a pharmacokinetic limitation for chronic metabolic disease applications, as it necessitates frequent dosing or formulation strategies to maintain therapeutic plasma concentrations. In the chronic metabolic disease studies, BAM15 was administered once daily by oral gavage at 100 mg/kg, producing daily exposure cycles rather than sustained plasma concentrations [3].

5.5 Routes of administration

Three routes have been used in preclinical studies: oral gavage (the principal route for chronic metabolic disease studies at doses of 50 to 100 mg/kg/day), intraperitoneal injection (used in acute pharmacokinetic studies and in sepsis/acute kidney injury models), and direct addition to drinking water or diet (used in some chronic studies) [1, 3, 6]. The oral route is the primary translational route. No parenteral, intravenous, or subcutaneous formulations have been described for BAM15 in the published literature.

5.6 Special populations

All published pharmacokinetic data are from rodent models (primarily C57BL/6J mice). No human pharmacokinetic data exist. Allometric scaling to human equivalent doses has not been formally published but can be estimated using standard body-surface-area scaling: the mouse dose of 100 mg/kg/day corresponds to an esti-

mated human equivalent dose of approximately 8 mg/kg/day (approximately 500 to 600 mg/day for a 70 kg human), though this estimate carries substantial uncertainty pending human pharmacokinetic characterization.

6. Preclinical Pharmacology

6.1 Diet-induced obesity

The Alexopoulos et al. (2020) study in *Nature Communications* is the foundational metabolic disease publication [3]. C57BL/6J mice fed a high-fat diet (60 percent kcal from fat) for 10 weeks and then treated with BAM15 at 100 mg/kg/day by oral gavage for an additional 5 weeks showed: (a) body weight reduction to near-chow-fed control levels; (b) fat mass reduction without loss of lean mass; (c) hepatic triglyceride reduction of approximately 75 percent; (d) reduction of hepatic non-esterified fatty acids; (e) decreased inflammatory lipid species in liver; (f) improved insulin sensitivity across liver, muscle, and adipose tissue as measured by hyperinsulinemic-euglycemic clamp; and (g) no change in food intake, body temperature, lean body mass, or hematological/biochemical safety markers [3].

Concurrently, Axelrod, Fealy, and colleagues (2020) demonstrated that BAM15 treatment of high-fat-diet-fed mice produced resistance to weight gain, improved body composition, improved glycemic control, enhanced mitochondrial respiratory kinetics *ex vivo*, and increased palmitate oxidation in liver tissue [2]. The AMPK-dependent mechanism was confirmed by *ex vivo* and *in vitro* experiments showing that BAM15 stimulated sustained AMPK phosphorylation and downstream substrate phosphorylation [2].

6.2 Head-to-head comparison with semaglutide and other agents

Axelrod et al. (2023) published a head-to-head comparison of five treatment interventions in female db/db mice (a genetic model of severe obesity, insulin resistance, and type 2 diabetes): calorie restriction (60 percent), semaglutide, rosiglitazone, BAM15, and nicosamide ethanolamine (NEN) [4]. BAM15 and calorie restriction improved body weight and liver steatosis to levels superior to semaglutide, NEN, and rosiglitazone. BAM15, semaglutide, and rosiglitazone completely restored glucose tolerance, while calorie restriction and NEN produced lesser improvements. BAM15-treated mice exhibited the most favorable lean-to-fat mass ratio among all treatment groups. All interventions improved hypertriglyceridemia [4].

6.3 Hepatic steatosis and NAFLD/MASH

BAM15 has demonstrated consistent hepatoprotective effects across multiple models of non-alcoholic fatty liver disease (NAFLD) and metabolic dysfunction-associated steatohepatitis (MASH). In high-fat-diet-fed mice, BAM15 reduced hepatic lipid accumulation by approximately 75 percent. Histologically, large vacuoles indicative of hepatosteatorosis in control animals were either small or absent in BAM15-treated animals [3]. In db/db mice, BAM15 reduced liver and serum triglyceride concentrations with accompanying reductions in ROS production and electron leakage [4].

Zhou et al. (2024) evaluated the combination of BAM15 with MGL-3196 (resmetirom), a thyroid hormone receptor beta agonist, in a mouse model of fatty liver disease. The combination had overall better efficacy than either monotherapy alone for improving metabolic outcomes [17].

6.4 Sepsis and acute kidney injury

Parikh, Kellum, and colleagues (2023) published a major study in the *Journal of Clinical Investigation* demonstrating that BAM15 treats mouse sepsis and sepsis-associated acute kidney injury [6]. In the cecal ligation and puncture (CLP) model of polymicrobial sepsis, intraperitoneal injection of BAM15 reduced mortality even when administered 12 hours after CLP, a time point at which mice were already clinically ill. BAM15 reduced circulating mitochondrial DNA (a damage-associated molecular pattern that drives systemic inflammation in sepsis), attenuated kidney tubule damage, reduced splenic apoptosis, improved systemic hemodynamics (reducing hypotension, increasing heart rate and body temperature), and decreased pro-inflammatory (IL-6) and anti-inflammatory (IL-10) systemic cytokines [6]. The mechanism was linked to reduced mitochondrial ROS production and consequent reduction in mitochondrial DNA release from damaged cells.

The original Kenwood et al. (2014) report also demonstrated that BAM15 dose-dependently protected against acute renal ischemia-reperfusion injury, providing the initial evidence for renal protection [1].

6.5 Acute myeloid leukemia

Gao et al. (2022) demonstrated that BAM15 significantly inhibited proliferation and promoted apoptosis in acute myeloid leukemia (AML) cell lines [7]. The mechanism was attributed to uncoupling-induced disruption of the mitochondrial membrane

potential, impairment of ATP synthesis, increased proton leakage and electron transport chain dysfunction, ROS accumulation, cytochrome c release, and caspase-mediated apoptosis. BAM15 was less cytotoxic to normal cells than to AML cells, reflecting the differential dependence of leukemic cells on oxidative phosphorylation. BAM15 was slightly superior to the classical uncouplers CCCP and FCCP in inhibiting tumor cell proliferation while sparing normal cells [7]. Subsequent research has extended these findings to non-small-cell lung cancer and T-cell malignancies.

6.6 Atherosclerosis and cardiovascular disease

Tai et al. (2018) demonstrated that BAM15 relaxed phenylephrine-constricted rat mesenteric arteries and potently activated AMPK in vascular smooth muscle cells (A10 cells) [5]. BAM15 was less cytotoxic in vascular smooth muscle cells compared with CCCP and niclosamide, and its AMPK-activating potency was significantly higher than that of metformin, AICAR, CCCP, and niclosamide [5].

Liu et al. (2025) extended these cardiovascular findings by demonstrating that oral BAM15 suppresses western diet-induced atherosclerosis in ApoE-knockout mice [11]. In macrophages, BAM15 suppressed NF-kappaB nuclear translocation and inhibited NLRP3 inflammasome expression and IL-1beta production. Both AMPK and NLRP3 are validated therapeutic targets for atherosclerosis, supporting the potential of BAM15 as a multi-target anti-atherosclerotic agent [11].

6.7 Sarcopenia and aging

Dantas et al. (2022) provided the first evidence that BAM15 prevents sarcopenic obesity in aged mice [8]. Mitochondrial uncoupling with BAM15 increased muscle mass and function, stimulated mitochondrial IL-6/STAT3 signaling, increased mitochondrial biogenesis and quality control through an AMPK-stimulated PGC-1alpha axis, increased oxidative phosphorylation activity, and reduced muscle endoplasmic reticulum stress, apoptotic signaling, and protein degradation [8]. The reduction in protein degradation was attributed to downregulation of FoxO3-stimulated transcription of atrogenes (MuRF1, atrogin-1) [8].

A 2025 bioRxiv preprint reported the first evaluation of BAM15 as a sarcopenia drug candidate in both male and female aged MitoQC mice [9]. BAM15 supplementation for eight weeks did not alter age-associated loss in muscle mass but pre-

vented a decline in skeletal muscle contractility and mitochondrial respiration, leading to functional improvements. The functional protection was associated with improved mitochondrial respiration and restoration of oxidative stress markers (4-HNE) in aged muscle [9].

7. Clinical Evidence Base

BAM15 has not entered human clinical trials. No Phase 1, Phase 2, or Phase 3 clinical data exist for any indication. The entire evidence base is preclinical, derived from cell culture experiments and rodent in vivo models (primarily C57BL/6J mice and db/db mice). This section documents the preclinical-to-clinical translational considerations.

7.1 Metabolic disease translation

The metabolic disease application (obesity, type 2 diabetes, NAFLD/MASH) represents the most developed translational pathway. The evidence base includes: reversal of established obesity in diet-induced and genetic mouse models; head-to-head superiority over semaglutide for body weight and hepatic steatosis; complete restoration of glucose tolerance; demonstrated safety at effective doses; oral bioavailability; and a defined molecular mechanism through AMPK and PGC-1alpha [2, 3, 4]. The principal translational barriers are: (a) the short plasma half-life (1.7 hours) which may necessitate extended-release formulations or more potent analogs for human dosing; (b) the high effective dose in mice (100 mg/kg/day, estimated human equivalent approximately 500 to 600 mg/day) which may limit formulation and tolerability; (c) the absence of human pharmacokinetic data; and (d) the absence of characterized metabolic pathways and drug-drug interaction profiles.

7.2 Sepsis and acute kidney injury translation

The JCI publication demonstrating efficacy 12 hours after sepsis onset is translational evidence of post-injury therapeutic utility, which is more clinically relevant than prophylactic dosing [6]. However, the parenteral route used in the sepsis studies (intraperitoneal injection) does not directly translate to human dosing, and an intravenous formulation would likely be required for critical care applications.

7.3 Oncology translation

The AML preclinical data demonstrate selective toxicity to leukemic cells, but the gap between in vitro concentrations and achievable in vivo plasma concentrations has not been bridged in published form [7]. Oncology translation would require formulation optimization and potentially combination strategies.

7.4 Comparison to other uncouplers in clinical development

Among mitochondrial uncouplers, only the controlled-release formulation of DNP (CRMP) and the oral antihelminthic niclosamide (repurposed for metabolic disease) have entered human clinical studies. HU6, a controlled-release DNP formulation developed by Bhatt and colleagues, is the most advanced mitochondrial uncoupler in clinical development, having completed early human trials. BAM15 itself has not advanced to clinical testing, though its improved safety profile relative to DNP in preclinical models supports the rationale for clinical translation [16].

8. Sourcing and Quality Verification

BAM15 is supplied as a research-grade compound by multiple chemical and pharmaceutical research suppliers. The compound is not available as a pharmaceutical-grade (GMP-manufactured) product, as it has not entered clinical development.

Commercial suppliers: Cayman Chemical (catalog number 17811), Sigma-Aldrich/MilliporeSigma (catalog number SML1760), Selleck Chemicals (catalog number S8535), MedChemExpress (MCE), Focus Biomolecules, BenchChem, and other research chemical vendors.

Typical purity: Greater than 95 percent by HPLC, with many suppliers providing material at greater than 98 percent purity. Certificate of analysis documents typically include HPLC purity, proton NMR spectrum, and mass spectrometry confirmation.

Pricing: Moderate for a research compound, approximately \$100 to \$400 per 50 mg depending on supplier and quantity. Gram quantities are available from some suppliers at reduced per-milligram cost.

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

- High-performance liquid chromatography area-percent purity, ideally greater than 98 percent
- Mass spectrometry confirmation of molecular weight (expected $[M+H]^+$ at m/z 341.10)
- Proton NMR confirmation of structure (characteristic aromatic proton pattern for 2-fluoroaniline substituents and N-H resonances)
- Assessment of residual solvents by gas chromatography (common synthesis solvents: THF, dichloromethane, ethyl acetate)

Third-party analytical laboratories (Janoshik Analytical, others) provide independent confirmation of vendor certificates of analysis. The research-grade BAM15 market is established and material from major suppliers is generally reliable, but lot-to-lot variability in purity can occur and analytical confirmation is appropriate for quantitative pharmacology studies.

9. Reconstitution and Handling

BAM15 is supplied as a solid powder (off-white to pale yellow crystalline material). The compound has low aqueous solubility and is typically handled as follows:

Stock solution preparation: Dissolve in DMSO at 10 to 50 mM concentration. For in vitro experiments, dilute DMSO stock solutions into cell culture medium immediately before use, keeping final DMSO concentration below 0.1 percent (v/v) to avoid solvent cytotoxicity. For in vivo oral gavage studies, BAM15 has been formulated in 15 percent Captisol (sulfobutylether-beta-cyclodextrin), which is the formulation vehicle used in the principal metabolic disease studies [3].

Storage: Store solid compound at -20 degrees Celsius, protected from light and moisture. DMSO stock solutions should be stored at -20 degrees Celsius and are stable for at least 6 months. Avoid repeated freeze-thaw cycles of stock solutions; aliquoting into single-use volumes is recommended.

Aqueous solubility: Very low in pure water (approximately 0.1 mg/mL or less). The low aqueous solubility necessitates solubilization vehicles (DMSO, cyclodextrin, other pharmaceutical excipients) for both in vitro and in vivo applications.

Handling precautions: Standard laboratory safety precautions for handling research chemicals. Avoid inhalation of powder and skin contact. Use in a fume hood when weighing. No specific hazardous decomposition products have been characterized, but the compound should be treated as pharmacologically active at low concentrations.

Stability: The solid compound is stable at room temperature under dry conditions for extended periods. Solutions in DMSO are stable at -20 degrees Celsius. Aqueous solutions and formulations should be used within 24 hours of preparation or stability-validated for longer storage.

10. Stack Interaction Considerations

BAM15 interacts with cellular energy metabolism at a fundamental level (the mitochondrial proton gradient), producing downstream effects on AMPK, mTOR, fatty acid oxidation, glucose uptake, and ROS production. Investigators conducting research with BAM15, especially in chronic dosing regimens or in combination with other compounds, should consider:

Other mitochondrial uncouplers: Concurrent use with DNP, FCCP, CCCP, niclosamide, or other protonophore uncouplers produces additive or synergistic dissipation of the mitochondrial proton gradient, increasing the risk of excessive uncoupling, ATP depletion, and cytotoxicity. The combination is not recommended unless specific dose-response characterization of the combination has been performed.

AMPK activators: Concurrent use with metformin, AICAR, or other AMPK-activating compounds produces additive AMPK activation. The combination of BAM15 and metformin may be pharmacologically productive for metabolic disease but has not been formally characterized. Additive effects on lactate production (from increased glycolysis) and on hepatic gluconeogenesis suppression should be monitored.

Thyroid hormone receptor agonists: The combination of BAM15 with resmetirom (MGL-3196) has been studied in a mouse model of fatty liver disease and showed better efficacy than either monotherapy alone [17]. This combination may be a productive research direction for NAFLD/MASH.

Insulin and insulin sensitizers: BAM15 improves insulin sensitivity through AMPK-mediated mechanisms. Concurrent use with insulin, thiazolidinediones (rosiglitazone, pioglitazone), or GLP-1 receptor agonists (semaglutide) may produce additive glycemic improvements but also additive hypoglycemia risk. The head-to-head comparison in db/db mice evaluated these agents in parallel arms rather than in combination [4].

Caloric restriction and fasting: The metabolic effects of BAM15 (increased substrate oxidation, fat loss, AMPK activation) overlap mechanistically with caloric restriction. The Axelrod et al. (2024) study demonstrated beneficial effects of simulta-

neously targeting calorie intake and calorie efficiency (BAM15) in diet-induced obese mice, suggesting that the combination may be productive [18].

Antioxidants: BAM15 reduces mitochondrial ROS production at mild uncoupling concentrations. Concurrent use with exogenous antioxidants (N-acetylcysteine, MitoQ, other mitochondria-targeted antioxidants) may produce additive antioxidant effects but could theoretically attenuate the ROS-mediated anticancer activity of BAM15 in oncology applications [7].

Substrates of hepatic CYP enzymes: The specific CYP isoforms responsible for BAM15 metabolism have not been identified. Concurrent use with known strong CYP inhibitors or inducers could alter BAM15 exposure, but this interaction has not been characterized. Investigators using BAM15 in combination with CYP-metabolized compounds should consider pharmacokinetic monitoring.

11. Adverse Events and Safety

11.1 Preclinical safety profile

The safety pharmacology of BAM15 in rodents has been notable for the absence of adverse findings at effective metabolic doses. In the Alexopoulos et al. (2020) study, C57BL/6J mice treated with BAM15 at 100 mg/kg/day for 5 weeks showed [3]:

- No change in body temperature (a critical safety parameter for mitochondrial uncouplers, as excessive uncoupling produces hyperthermia; DNP-related deaths in humans were attributed to uncontrolled hyperthermia)
- No change in food intake
- No change in lean body mass (fat loss was selective)
- No change in standard hematological parameters (complete blood count)
- No change in standard biochemical parameters (liver enzymes, renal function markers, electrolytes)
- No change in physical activity or behavioral parameters

11.2 Comparison to classical uncoupler toxicity

The safety advantage of BAM15 over classical uncouplers is substantial. 2,4-Dinitrophenol was marketed as an obesity drug in the 1930s and was withdrawn by the FDA in 1938 following reports of fatal hyperthermia, cataracts, agranulocytosis, and peripheral neuropathy [16]. The therapeutic index of DNP is narrow; the difference between an effective dose and a lethal dose is small, and inter-individual variability in metabolic rate and ambient temperature further narrows the margin. BAM15 addresses this limitation through mitochondrial membrane selectivity, avoiding the plasma membrane depolarization and resultant cytotoxicity that contribute to DNP and FCCP toxicity [1, 2].

In cell culture, BAM15 did not induce caspase 3/7 activation at concentrations up to 40 micromolar, while DNP and FCCP induced caspases at all tested concentrations [1]. In vascular smooth muscle cells, BAM15 was less cytotoxic than CCCP and niclosamide [5].

11.3 Potential safety concerns

Despite the favorable preclinical safety profile, several potential safety concerns should be considered for future translational development:

- **Hyperthermia risk at high doses:** While body temperature was unaffected at 100 mg/kg/day in mice, excessive uncoupling at supratherapeutic doses would be expected to produce heat generation exceeding thermoregulatory capacity. The dose-response relationship for hyperthermia has not been fully characterized.
- **Hepatic effects:** Primary liver distribution means that the highest tissue concentrations are in the liver. While no hepatotoxicity was observed at studied doses, chronic high-dose exposure could potentially produce hepatic mitochondrial dysfunction. Hepatic safety monitoring would be essential in clinical development.
- **Cardiac effects:** Mitochondrial uncoupling in cardiomyocytes could theoretically impair cardiac bioenergetics. No cardiac adverse effects have been reported in the published preclinical literature, but formal cardiac safety pharmacology (hERG channel assay, telemetry in conscious animals) has not been published.
- **Reproductive and developmental toxicity:** No reproductive or developmental toxicity studies have been published.
- **Long-term carcinogenicity:** No carcinogenicity studies have been published.

11.4 Therapeutic index

The preclinical therapeutic index of BAM15 is favorable relative to classical uncouplers. The effective metabolic dose (100 mg/kg/day oral in mice) is well below doses that produce observable toxicity. The exact lethal dose (LD50) has not been published, but the absence of toxicity at the studied dose levels suggests a therapeutic index substantially wider than that of DNP or FCCP.

12. Comparable Compounds Assessment

This section presents a structured comparative assessment of five alternative mitochondrial uncouplers against BAM15, evaluated on five standards: novelty, effect size, promising potential (current development trajectory), side-effect profile, and overall validation. The five comparators were selected as the principal mitochondrial uncouplers with substantive preclinical or clinical pharmacology programs, with priority given to compounds that share BAM15's metabolic disease positioning.

12.1 2,4-Dinitrophenol (DNP)

Mechanism: Non-selective protonophore uncoupler that dissipates proton gradients across both the inner mitochondrial membrane and the plasma membrane. The prototypical mitochondrial uncoupler.

Development trajectory: DNP was marketed as an obesity drug in the United States from 1933 to 1938, producing weight loss of 1 to 1.5 kg per week at typical doses. The FDA withdrew the drug following reports of fatal hyperthermia, cataracts, agranulocytosis, and peripheral neuropathy [16]. Modern interest has centered on controlled-release formulations (CRMP/HU6) designed to achieve low, sustained plasma concentrations that produce mild uncoupling without the dose spikes that contributed to historical toxicity. Perry et al. (2015) demonstrated that controlled-release DNP reversed hepatic steatosis and insulin resistance in rats at plasma concentrations approximately 100-fold below the toxic threshold [19].

Novelty: Low. DNP is the oldest mitochondrial uncoupler and the historical reference compound.

Effect size: Substantial for weight loss and metabolic improvement, but the therapeutic window is narrow with the immediate-release formulation. The controlled-release formulation achieves metabolic effects at safer plasma concentrations.

Promising potential: Moderate. The HU6 controlled-release DNP formulation is the most clinically advanced mitochondrial uncoupler and has entered early human testing. The parent compound is not viable for development.

Side-effect profile: The immediate-release formulation has an unacceptable safety profile: fatal hyperthermia, cataracts with chronic use, agranulocytosis, peripheral neuropathy, and diaphoresis. The controlled-release formulation appears safer at reduced plasma concentrations, but human safety data remain limited.

Overall validation: High for proof of concept (metabolic effects are well established over 90 years of research); low for clinical viability of the parent compound.

Comparison to BAM15: BAM15 was specifically designed to address the fundamental safety limitation of DNP, namely non-selective membrane depolarization. BAM15 achieves comparable or superior metabolic effects in rodent models without the plasma membrane toxicity, without hyperthermia, and without the adverse safety markers that preclude DNP development. The DNP controlled-release strategy is an alternative approach to the same problem (achieving mild uncoupling without toxicity) but addresses it through formulation rather than through molecular selectivity.

12.2 FCCP (Carbonylcyanide-p-trifluoromethoxyphenylhydrazine)

Mechanism: Non-selective protonophore uncoupler of equal potency to BAM15 in mitochondrial uncoupling assays. Widely used as a research tool for maximal mitochondrial respiration measurements (the "FCCP-stimulated respiration" parameter in Seahorse and Oroboros assays).

Development trajectory: FCCP is a laboratory research tool and has never been developed for therapeutic applications owing to its narrow effective range and cytotoxicity at concentrations only modestly above the uncoupling threshold.

Novelty: Low. FCCP is the standard laboratory uncoupler reference compound.

Effect size: Potent mitochondrial uncoupling in vitro. In vivo utility is limited by cytotoxicity.

Promising potential: None as a therapeutic candidate. Indefinite utility as a research tool.

Side-effect profile: FCCP begins to inhibit oxygen consumption rate at 1 to 2 micromolar (reflecting cytotoxicity from plasma membrane depolarization), whereas BAM15 maintains stimulation up to 8 micromolar [1]. FCCP induces caspase 3/7 activity at all tested concentrations. The compound is unsuitable for in vivo therapeutic application.

Overall validation: High as a research tool; not applicable as a therapeutic candidate.

Comparison to BAM15: BAM15 was identified through a screen designed to find a superior alternative to FCCP. BAM15 matches FCCP potency in mitochondrial uncoupling but produces higher maximal respiration rates, longer-duration respiratory stimulation, and dramatically reduced cytotoxicity. BAM15 is the functional successor to FCCP for applications requiring in vivo efficacy.

12.3 Niclosamide

Mechanism: Anthelmintic drug (FDA-approved for tapeworm infections) that also functions as a mitochondrial uncoupler through protonophore activity. Repurposed for metabolic disease applications as niclosamide ethanolamine (NEN), a salt form with improved oral bioavailability [20].

Development trajectory: NEN has been evaluated in preclinical models of obesity and diabetes. Tao et al. (2014) demonstrated that NEN treatment of high-fat-diet-fed mice reduced weight gain and improved glycemic control [20]. The Axelrod et al. (2023) head-to-head comparison in db/db mice showed that NEN was inferior to BAM15, semaglutide, and calorie restriction for body weight reduction and glucose tolerance improvement [4].

Novelty: Low to moderate. The uncoupling mechanism of an established drug is a repurposing strategy rather than a novel discovery.

Effect size: Modest in preclinical metabolic disease models, inferior to BAM15 in the head-to-head comparison.

Promising potential: Low to moderate. The compound is FDA-approved for an unrelated indication, which could facilitate clinical repositioning, but the metabolic disease effect size is modest and the cytotoxicity profile is less favorable than BAM15.

Side-effect profile: Niclosamide is more cytotoxic than BAM15 in vascular smooth muscle cells [5]. The compound has known gastrointestinal adverse effects at antihelminthic doses. The uncoupling mechanism operates across both mitochondrial and plasma membranes, similar to DNP and FCCP, without BAM15's mitochondrial selectivity.

Overall validation: Moderate. Established safety profile at antihelminthic doses; limited metabolic disease validation.

Comparison to BAM15: BAM15 is superior to NEN for metabolic endpoints in the head-to-head db/db comparison [4] and demonstrates lower cytotoxicity and higher AMPK activation potency [5]. NEN has the advantage of existing FDA approval (for an unrelated indication) and established human safety data at antihelminthic doses. BAM15 has the advantage of mitochondrial selectivity and stronger preclinical metabolic efficacy.

12.4 OPC-163493

Mechanism: Mitochondrial uncoupler developed by Otsuka Pharmaceutical through a medicinal chemistry program designed to identify safe, orally bioavailable uncouplers. The compound was identified through a cell-based screen similar in concept to the BAM15 discovery screen [21].

Development trajectory: OPC-163493 was characterized in preclinical obesity and diabetes models. Kanemoto et al. (2019) demonstrated that OPC-163493 decreased plasma glucose, hepatic steatosis, and body weight in multiple rodent models of diabetes (Zucker diabetic fatty rats, OLETF rats, and db/db mice) without affecting body temperature or food intake [21]. The compound has a longer plasma half-life than BAM15 (approximately 4 hours in rats), which represents a pharmacokinetic advantage.

Novelty: Moderate. The compound represents independent parallel discovery of a safe mitochondrial uncoupler from a pharmaceutical company program.

Effect size: Comparable to BAM15 in glucose-lowering and anti-steatosis effects in rodent models. Head-to-head comparison with BAM15 has not been published.

Promising potential: Moderate. Pharmaceutical company backing (Otsuka) provides development resources, but clinical advancement has not been publicly reported.

Side-effect profile: Favorable preclinical safety profile similar to BAM15: no hyperthermia, no effect on food intake, no hematological or biochemical toxicity at effective doses [21].

Overall validation: Moderate. Developed through a systematic pharmaceutical program with multiple rodent model studies.

Comparison to BAM15: OPC-163493 and BAM15 represent parallel approaches to the same pharmacological objective (safe mitochondrial uncoupling for metabolic disease). OPC-163493 has a potentially longer half-life, which is a pharmacokinetic advantage. BAM15 has a broader preclinical evidence base spanning metabolic disease, sepsis, cancer, atherosclerosis, and sarcopenia, whereas OPC-163493 characterization has been more narrowly focused on metabolic endpoints. Both compounds share the critical safety feature of absent hyperthermia at effective doses.

12.5 SHM115

Mechanism: Oxadiazolopyridine derivative developed through structure-activity optimization of the BAM15 scaffold. SHM115 retains the protonophore uncoupling mechanism of BAM15 but incorporates structural modifications (substitution of one aniline group with a hydroxyl moiety and migration from the pyrazine to the pyridine core) designed to improve tissue distribution to adipose tissue [15].

Development trajectory: SHM115 was reported by the Webster-Santos laboratory as a second-generation BAM15 analog with improved distribution to white and brown adipose tissue. The compound demonstrated efficacy in diet-induced obesity prevention in rodent models [15].

Novelty: Moderate. SHM115 is an optimized derivative rather than a novel scaffold, but the tissue distribution improvement represents a meaningful pharmacological advance over BAM15.

Effect size: Comparable to BAM15 in metabolic disease models, with potentially improved adipose tissue effects owing to the superior tissue distribution.

Promising potential: Moderate. As a direct successor to BAM15, SHM115 may represent the lead development candidate for clinical translation. Published data are limited relative to the extensive BAM15 literature.

Side-effect profile: Limited published data. Expected to retain the favorable safety profile of BAM15 given the conserved mechanism and related scaffold.

Overall validation: Low to moderate. The compound is early in characterization relative to BAM15.

Comparison to BAM15: SHM115 is a purpose-built optimization of BAM15 designed to address the limitation of modest adipose tissue distribution. If validated, SHM115 would supersede BAM15 for metabolic disease applications targeting adipose tissue metabolism. BAM15 retains the advantage of a substantially larger evidence base across multiple disease models and the longest track record among the safe mitochondrial uncouplers.

12.6 Comparator summary

The five comparators span the history and future of mitochondrial uncoupler pharmacology. **DNP** is the historical prototype, effective but unacceptably toxic in its conventional form, with renewed interest through controlled-release formulations. **FCCP** is the standard research tool, potent but unsuitable for in vivo therapy. **Niclosamide/NEN** is a repositioned antihelminthic with modest metabolic effects inferior to BAM15 in head-to-head comparison. **OPC-163493** is a parallel pharmaceutical discovery with similar safety profile but narrower indication development. **SHM115** is a purpose-built BAM15 successor with improved tissue distribution and early-stage characterization.

BAM15 occupies a central position in this landscape: it is the most extensively characterized safe mitochondrial uncoupler across the broadest range of disease indications (metabolic disease, sepsis, cancer, atherosclerosis, sarcopenia); it has the largest published preclinical evidence base; and it has demonstrated the critical safety features (mitochondrial selectivity, absent hyperthermia, no lean mass loss, preserved food intake) that distinguish it from the classical uncouplers. Its principal limitations are the short plasma half-life (1.7 hours), the high effective dose in rodents, and the absence of human clinical data. These limitations are the focus of the ongoing medicinal chemistry optimization programs (SHM115, other analogs) and formulation development that constitute the translational bridge to clinical testing.

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