

RESEARCH MONOGRAPH . COMPOUND PROFILE

Pinealon

A synthetic tripeptide bioregulator derived from the pineal gland peptide fraction, investigated as a neuroepigenetic modulator of gene expression with reported neuroprotective, antioxidant, and geroprotective activity in preclinical oxidative stress, ischemia, and neurodegeneration models.

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ABSTRACT

Pinealon (EDR peptide; L-glutamyl-L-aspartyl-L-arginine) is a synthetic tripeptide bioregulator developed at the Saint Petersburg Institute of Bioregulation and Gerontology under the direction of Vladimir Khavinson as part of a multigenerational program to identify tissue-specific short-chain peptides capable of modulating gene expression through direct interaction with chromatin. The compound was identified as one of the shortest biologically active sequences within Cortexin, a complex peptide fraction extracted from bovine cerebral cortex tissue that has been used clinically in several post-Soviet jurisdictions for the treatment of traumatic brain injury, ischemic stroke, and cognitive impairment. Pinealon was subsequently synthesized as a standalone tripeptide and advanced through a series of *in vitro* and *in vivo* investigations spanning antioxidant activity, neuroprotection, serotonin biosynthesis regulation, dendritic spine preservation in Alzheimer's disease models, and preliminary open-label clinical observations in elderly patients with cognitive decline and in patients recovering from craniocerebral trauma.

The proposed mechanism of action is unconventional relative to classical receptor-mediated peptide pharmacology. Due to its low molecular weight (418.41 g/mol) and cationic character, Pinealon is reported to penetrate lipid bilayers and nuclear membranes without requiring surface receptor engagement, gaining direct access to chromatin. Molecular modeling and *in vitro* binding studies from the Khavinson laboratory have identified complementary binding sites in the promoter regions of several genes relevant to neuroprotection and neurodegeneration,

including TPH1 (tryptophan hydroxylase 1, the rate-limiting enzyme in serotonin biosynthesis), SOD2 (mitochondrial superoxide dismutase), GPX1 (glutathione peroxidase 1), PPARA and PPARG (peroxisome proliferator-activated receptor alpha and gamma), CASP3 (caspase-3), and APOE (apolipoprotein E). The proposed binding occurs at specific DNA sequences, principally d(CCTGCC)₂ and d(CCAGC)₂, through sequence-specific steric and electrostatic complementarity with the major groove of double-stranded DNA. The functional consequence is reported to be destabilization of local DNA secondary structure, alteration of histone modification patterns, and increased accessibility of regulatory regions to transcription factors, resulting in upregulation of neuroprotective gene products and downregulation of pro-apoptotic pathways.

Preclinical pharmacology studies, conducted predominantly by the Khavinson group and affiliated Russian laboratories, have reported that Pinealon produces dose-dependent suppression of reactive oxygen species accumulation in cerebellar granule cells, neutrophils, and pheochromocytoma (PC12) cells; increases cell viability under oxidative stress conditions; delays ERK1/2 activation in neurons exposed to homocysteine; reduces caspase-3 expression and p53 protein synthesis in brain tissue; increases serotonin synthesis in neuronal cultures of rat cerebral cortex; normalizes superoxide dismutase and glutathione peroxidase activity in the brains of hypoxia-sensitive rats; prevents the loss of mushroom-shaped dendritic spines in hippocampal neurons from 5xFAD transgenic mice (a model of familial Alzheimer's disease); and protects rat offspring from prenatal hyperhomocysteinemia-induced cognitive deficits. In an open-label clinical observation in 72 patients with traumatic brain injury, addition of Pinealon to

standard rehabilitation therapy improved memory function in approximately 59 percent of patients.

No completed, peer-reviewed randomized controlled trial of Pinealon has been published in English-language indexed journals as of the date of this monograph. No Phase 1 formal safety study, no Phase 2 efficacy trial, and no Phase 3 registration study exist in any population. The compound has no approved indication in any jurisdiction recognized by the International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use. Virtually all published Pinealon research originates from the Khavinson laboratory and closely affiliated institutions; independent replication by Western academic laboratories is absent. The chromatin-interaction model, while supported by computational molecular modeling and fluorescence microscopy studies from the originating group, has not been independently validated by structural biology methods (X-ray crystallography, cryo-electron microscopy) at the resolution required to confirm the proposed binding geometry. This monograph documents the chemistry, proposed mechanism, preclinical pharmacology, clinical observations, sourcing and handling, and comparative assessment of the compound, and identifies the principal evidence gaps that currently limit its positioning in the research-clinical translation pipeline.

1. Compound Identification

1.1 Names, synonyms, and registry identifiers

Common name: Pinealon.

Peptide sequence designation: EDR (single-letter amino acid code); Glu-Asp-Arg (three-letter code).

Full chemical name: L-glutamyl-L-aspartyl-L-arginine.

Chemical Abstracts Service registry number: 175175-23-2.

PubChem Compound Identifier: CID 10273502.

Development context: The compound was developed at the Saint Petersburg Institute of Bioregulation and Gerontology (Saint Petersburg, Russian Federation) as a second-generation synthetic bioregulatory peptide ("cytogen") derived from the biologically active fraction of bovine pineal gland and cerebral cortex tissue. The name "Pinealon" reflects the pineal gland origin of the parent peptide fraction from which the EDR sequence was identified as a minimal active unit [1, 2].

Related compounds: Cortexin (the complex peptide fraction from bovine cerebral cortex containing EDR among other sequences); Epithalon (AEDG tetrapeptide, another Khavinson-group pineal bioregulator targeting telomerase); KED (Lys-Glu-Asp, a related Khavinson tripeptide with overlapping neuroprotective activity) [3].

1.2 Structural class and chemistry

Pinealon is a linear, unmodified tripeptide consisting of three proteinogenic L-amino acids linked by standard peptide bonds: L-glutamic acid at the amino terminus, L-aspartic acid at the central position, and L-arginine at the carboxy terminus.

Molecular formula: C₁₅H₂₆N₆O₈.

Molecular weight: 418.41 g/mol (monoisotopic exact mass: 418.1813 g/mol).

Physical properties: White to off-white lyophilized powder. Freely soluble in water and buffered aqueous solutions at physiological pH. Soluble in dimethyl sulfoxide. The compound carries a net positive charge at physiological pH owing to the guanidinium group of the arginine residue (pKa approximately 12.5), which remains protonated under all biologically relevant conditions. The two acidic residues (glutamic acid, pKa approximately 4.1; aspartic acid, pKa approximately 3.9) are deprotonated at physiological pH, contributing two negative charges. The net charge at pH 7.4 is therefore approximately zero to slightly positive depending on the protonation state of the terminal amino and carboxyl groups. The cationic character of the arginine guanidinium is proposed as a critical feature enabling electrostatic interaction with the negatively charged phosphodiester backbone of DNA in the major groove [4, 5].

Stereochemistry: All three amino acid residues are in the L-configuration. No D-amino acid variants have been reported in the Khavinson literature. Investigators acquiring research-grade Pinealon should confirm chirality by chiral chromatography or by comparison of optical rotation against a reference standard of defined stereochemistry.

1.3 Synthesis

Pinealon is synthesized by standard solid-phase peptide synthesis (Fmoc chemistry) or by solution-phase coupling. The synthesis is straightforward: the three amino acids are sequentially coupled using standard carbodiimide or uronium-based activating reagents (HBTU, HATU, or DIC/HOBt), with orthogonal side-chain protecting groups for the glutamic acid gamma-carboxyl (typically OtBu), the aspartic acid beta-carboxyl (OtBu), and the arginine guanidinium (Pbf or Pmc). Global deprotection and cleavage from resin (for solid-phase synthesis) is accomplished with trifluoroacetic acid cocktails. The crude peptide is purified by reversed-phase high-performance liquid chromatography and characterized by mass spectrometry and analytical HPLC. Yields are typically high (greater than 80 percent crude, greater than 95 percent after purification) owing to the short chain length and absence of difficult coupling steps. The compound is supplied as the trifluoroacetate salt or as the acetate salt after counterion exchange; the acetate salt is preferred for biological applications to avoid potential trifluoroacetate interference in cell-based assays [6].

2. Abstract

Pinealon (EDR; L-glutamyl-L-aspartyl-L-arginine) is a synthetic tripeptide bioregulator developed by the Khavinson group at the Saint Petersburg Institute of Bioregulation and Gerontology as part of a systematic program to identify ultra-short peptide sequences capable of tissue-specific gene expression modulation through direct chromatin interaction [1, 2]. The compound was identified as one of the minimal biologically active sequences within Cortexin, a complex neuropeptide fraction extracted from bovine cerebral cortex that has been used clinically in the Russian Federation and several post-Soviet states for the treatment of ischemic stroke, traumatic brain injury, and cognitive impairment in pediatric and adult populations.

The proposed mechanism of action diverges fundamentally from classical receptor-mediated peptide signaling. Rather than binding extracellular or intracellular receptors, Pinealon is reported to penetrate cellular and nuclear membranes by virtue of its small size and cationic arginine residue, gaining direct access to chromatin. The Khavinson laboratory has published molecular modeling, fluorescence microscopy, and in vitro DNA binding studies proposing that the tripeptide binds specific nucleotide sequences in promoter regions of neuroprotection-relevant genes through complementary electrostatic and steric interactions with the DNA major groove [4, 5]. The proposed target genes include TPH1 (tryptophan hydroxylase 1), SOD2 (superoxide dismutase 2), GPX1 (glutathione peroxidase 1), PPARA and PPARG (peroxisome proliferator-activated receptors), CASP3 (caspase-3), NES (nestin), GAP43 (growth-associated protein 43), and APOE (apolipoprotein E) [5, 7].

Preclinical evidence, derived almost exclusively from the Khavinson group and affiliated laboratories, supports antioxidant activity (dose-dependent suppression of reactive oxygen species in cerebellar granule cells, neutrophils, and PC12 cells), neuroprotection (reduced caspase-3 expression, delayed ERK1/2 activation under excitotoxic conditions, preservation of dendritic spine morphology in Alzheimer's disease models), serotonin biosynthesis upregulation (increased tryptophan hydroxylase expression in cortical neuron cultures), and cognitive protection in rodent models of prenatal hyperhomocysteinemia and experimentally induced diabetes [8, 9, 10, 11, 12].

Clinical evidence is limited to open-label observational studies without randomization or blinding. In an observation of 72 patients with traumatic brain injury receiving Pinealon as an adjunct to standard rehabilitation, memory improvement was reported in approximately 59 percent of subjects [13]. No randomized controlled trial, no formal Phase 1 safety study, and no registration trial has been published for Pinealon in any population or indication. The compound has no approved clinical indication in any jurisdiction. Independent replication of the core mechanistic and pharmacological claims by laboratories outside the Khavinson network is absent as of the date of this monograph.

3. Discovery and Development History

3.1 Origin within the Khavinson bioregulator program

The discovery of Pinealon is inseparable from the broader bioregulatory peptide research program initiated by Vladimir Khavinson at the Military Medical Academy in Leningrad (now Saint Petersburg) in the late 1970s and continued at the Saint Petersburg Institute of Bioregulation and Gerontology from the early 1990s onward. The foundational hypothesis of this program is that short-chain peptides (2 to 7 amino acids), isolated from specific organ tissues or synthesized de novo, can modulate gene expression in a tissue-specific manner by direct interaction with chromatin, and that this modulation can restore age-related or pathology-related declines in organ function [1, 2].

The first generation of Khavinson compounds ("cytomaxes") consisted of complex peptide fractions extracted from animal organs by acid hydrolysis and ultrafiltration. Cortexin, the cerebral cortex-derived cytomaxe, was developed in the 1980s and received regulatory approval in the Russian Federation for the treatment of traumatic brain injury, ischemic stroke, viral encephalitis, epilepsy, and cognitive impairment. Cortexin is a polypeptide mixture containing multiple sequences; the identification of individual active components within this mixture was the basis for the second-generation program [14].

3.2 Identification of the EDR sequence

The second generation of Khavinson compounds ("cytogens") consisted of synthetic short-chain peptides with defined amino acid sequences, identified by fractionation and bioactivity-guided isolation from the parent cytomaxe preparations. Pinealon (EDR; Glu-Asp-Arg) was identified as one of the minimal biologically active sequences within the Cortexin fraction and, independently, within peptide fractions derived from bovine pineal gland tissue. The name "Pinealon" reflects this pineal gland association, although the compound's principal investigated activities are neuronal rather than pineal-endocrine [1, 2].

The identification process, as described in the Khavinson group's publications, involved extraction of peptide fractions from bovine pineal and cortical tissue, purification by sequential ultrafiltration and chromatographic separation, sequencing of biologically active fractions, and synthesis of candidate minimal sequences for testing in cell-based neuroprotection assays. The EDR tripeptide was selected for further development based on its activity in oxidative stress protection and cell viability assays in neuronal cultures [8].

3.3 Development of Pinealon as a standalone compound

Following identification of the EDR sequence, the Khavinson group synthesized Pinealon as a standalone compound and conducted a series of in vitro and in vivo investigations beginning in approximately 2005 and continuing through the present. The principal lines of investigation have included antioxidant activity in neuronal cell cultures, neuroprotection in ischemia and excitotoxicity models, gene expression modulation (particularly of tryptophan hydroxylase and antioxidant enzymes), DNA binding characterization by molecular modeling and biophysical methods, dendritic spine preservation in Alzheimer's disease models, and open-label clinical observations in traumatic brain injury and age-related cognitive decline [5, 8, 9, 10, 11, 12, 13].

Pinealon has been commercialized in the Russian Federation as a dietary supplement (capsule formulation) for cognitive support and has been marketed through Russian and international peptide supplement distributors. The compound has not undergone formal regulatory review as a pharmaceutical in any jurisdiction recognized by the International Council for Harmonisation.

3.4 Current research-clinical status

As of the date of this monograph, Pinealon occupies a position of active preclinical investigation with preliminary clinical observations but no formal clinical development pathway. The published evidence base consists entirely of work from the Khavinson group and closely affiliated Russian institutions. No independent Western laboratory has published a replication study of the core pharmacological or mechanistic claims. No investigational new drug application, no clinical trial registration in ClinicalTrials.gov, and no regulatory submission for Pinealon has been identified in publicly accessible registries.

4. Mechanism of Action

4.1 Proposed epigenetic mechanism: direct chromatin interaction

The Khavinson laboratory has proposed a mechanism of action for Pinealon (and for other ultrashort bioregulatory peptides) that is fundamentally distinct from conventional receptor-mediated peptide signaling. The central claim is that tripeptides and dipeptides of appropriate size, charge, and sequence can penetrate cell membranes and nuclear membranes without receptor mediation, enter the nucleus, and interact directly with chromatin to modulate gene expression [4, 5].

The physicochemical basis for the proposed membrane penetration is the small molecular size (418.41 g/mol, well below the conventional 500 Da threshold for passive membrane permeability) and the cationic character conferred by the arginine guanidinium group, which facilitates electrostatic interaction with negatively charged phospholipid headgroups. The Khavinson group has published fluorescence microscopy studies reporting visualization of labeled short peptides within cell nuclei and nucleoli following extracellular application, supporting the claim of nuclear access [4].

Within the nucleus, the proposed interaction is binding to specific DNA sequences in the major groove of double-stranded DNA. Molecular modeling studies from the Khavinson laboratory have identified two candidate binding sequences for the EDR tripeptide: d(CCTGCC)₂ and d(CCAGC)₂ [5]. The binding is proposed to occur through complementary electrostatic interactions between the charged amino acid side chains and the nucleotide bases and phosphodiester backbone, with the guanidinium group of arginine forming hydrogen bonds with guanine bases in the major groove, and the carboxylate groups of glutamic acid and aspartic acid interacting with the sugar-phosphate backbone. In vitro biophysical studies (circular dichroism, viscometry) have reported that the EDR peptide exerts a destabilizing effect on the secondary structure of double-stranded DNA while compacting its molecular coil, consistent with a local perturbation of DNA conformation at the binding sites [5].

The functional consequence of this DNA interaction is proposed to be alteration of chromatin accessibility. By binding promoter regions and perturbing local DNA conformation, the peptide is hypothesized to facilitate or inhibit the binding of transcription factors and RNA polymerase, thereby modulating the expression of downstream genes. The Khavinson group has additionally proposed that short peptides interact with histone proteins within the nucleosome complex, altering histone modification patterns (acetylation, methylation) and further modulating chromatin accessibility [4, 15].

4.2 Target genes and downstream pathways

Molecular modeling and gene expression studies from the Khavinson laboratory have identified promoter-region binding sites for the EDR peptide in the following genes [5, 7]:

TPH1 (tryptophan hydroxylase 1): One binding site (CCTGCC sequence). TPH1 encodes the rate-limiting enzyme in the biosynthesis of serotonin from tryptophan. Upregulation of TPH1 expression by EDR is proposed as the molecular basis for the observed increase in serotonin synthesis in cortical neuron cultures and for downstream effects on melatonin biosynthesis through the tryptophan-serotonin-melatonin pathway [9, 16].

SOD2 (superoxide dismutase 2, mitochondrial): Binding site identified. SOD2 encodes the mitochondrial manganese superoxide dismutase, the principal enzymatic defense against superoxide radical generation in the mitochondrial matrix. Upregulation of SOD2 is proposed as a contributor to the antioxidant neuroprotective activity of Pinealon [5, 8].

GPX1 (glutathione peroxidase 1): Binding site identified. GPX1 encodes a selenocysteine-containing enzyme that catalyzes the reduction of hydrogen peroxide and organic hydroperoxides by glutathione. Coordinate upregulation of SOD2 and GPX1 is proposed to normalize the antioxidant defense system in hypoxia-sensitive neurons [5, 8].

PPARA and PPARG (peroxisome proliferator-activated receptors alpha and gamma): Three and five binding sites, respectively. These nuclear receptors regulate lipid metabolism, inflammation, and cell differentiation. PPARG activation in microglia has been associated with anti-inflammatory neuroprotection in multiple

neurodegeneration models. The significance of the multiple binding sites in PPARG is not fully elucidated but may indicate a particularly strong regulatory interaction [5].

CASP3 (caspase-3): Binding site identified. Caspase-3 is the principal executioner caspase in apoptotic cell death. Downregulation of CASP3 expression is proposed as a direct anti-apoptotic mechanism contributing to the neuroprotective effect of Pinealon in ischemia and excitotoxicity models [5, 7, 10].

APOE (apolipoprotein E): Binding site identified. APOE is the principal cholesterol transport protein in the central nervous system and the strongest common genetic risk factor for late-onset Alzheimer's disease. The functional significance of EDR binding to the APOE promoter has not been characterized beyond the identification of the binding site [7].

NES (nestin) and GAP43 (growth-associated protein 43): Binding sites identified. Both genes encode proteins associated with neuronal growth, differentiation, and plasticity. Upregulation is proposed to support neuroplasticity and neuronal repair under pathological conditions [7].

4.3 MAPK/ERK pathway modulation

The Khavinson group has reported that Pinealon modulates the mitogen-activated protein kinase (MAPK) / extracellular signal-regulated kinase (ERK) signaling pathway in neurons exposed to excitotoxic insults. In cerebellar granule cells exposed to homocysteine (an endogenous excitotoxin implicated in neurodegeneration), EDR peptide treatment delayed ERK1/2 activation, with phosphorylation appearing at 20 minutes versus 2.5 minutes in untreated controls [5]. The delay in ERK1/2 activation is interpreted as a protective modulation of the stress-response signaling cascade, preventing the rapid, maladaptive activation pattern associated with excitotoxic neuronal death while preserving the slower, adaptive activation pattern associated with cell survival and plasticity.

4.4 Serotonin biosynthesis regulation

A distinctive reported activity of Pinealon is the upregulation of serotonin synthesis in neuronal cultures of rat cerebral cortex. The Khavinson group has published studies demonstrating that EDR peptide treatment increased tryptophan hydroxylase (TPH1) expression and serotonin levels in cortical neuron cultures [9, 16]. The

CCTGCC nucleotide sequence in the TPH1 gene promoter was identified as complementary to the EDR peptide, and the serotonin-stimulatory activity was attributed to epigenetic upregulation of this gene. This finding positions Pinealon within the broader tryptophan-serotonin-melatonin biosynthetic axis and provides a proposed molecular basis for reported effects on circadian rhythm modulation and sleep-wake cycle regulation [16].

4.5 Limitations of the proposed mechanism

The chromatin-interaction model for Pinealon's mechanism of action, while internally consistent within the Khavinson group's publications, has not been validated by independent laboratories using orthogonal structural biology methods. Specific limitations include: (a) the absence of high-resolution structural data (X-ray crystallography, cryo-electron microscopy, or nuclear magnetic resonance spectroscopy) confirming the proposed peptide-DNA binding geometry at atomic resolution; (b) the absence of chromatin immunoprecipitation followed by sequencing (ChIP-seq) data confirming genomic binding sites in living cells; (c) the absence of independent replication of the fluorescence microscopy studies reporting nuclear peptide localization; (d) the thermodynamic plausibility of a tripeptide achieving sequence-specific DNA binding at the proposed concentrations, given the relatively weak electrostatic interactions available to a molecule of this size; and (e) the absence of dose-response pharmacodynamic studies correlating intranuclear peptide concentration with gene expression changes in a quantitative, mechanistically rigorous framework [17].

These limitations do not invalidate the published observations of biological activity but indicate that the mechanistic interpretation remains a hypothesis supported by circumstantial evidence rather than a proven molecular mechanism.

5. Pharmacokinetics and Routes of Administration

5.1 Absorption and bioavailability

Formal pharmacokinetic studies of Pinealon in humans have not been published in peer-reviewed literature. The pharmacokinetic profile is inferred from general principles of tripeptide absorption and from the limited animal data available in the Khavinson group's publications.

Oral bioavailability of unmodified tripeptides is generally low owing to rapid enzymatic degradation by gastric, pancreatic, and brush-border peptidases and to limited passive permeability across the intestinal epithelium. However, di- and tripeptides benefit from active transport by the intestinal peptide transporter PEPT1 (SLC15A1), which mediates the proton-coupled uptake of small peptides across the apical membrane of enterocytes. The extent to which Pinealon is a substrate for PEPT1 has not been formally characterized, but the tripeptide structure is consistent with transporter recognition [18].

Sublingual administration has been proposed as an alternative route to bypass first-pass gastrointestinal degradation, and sublingual formulations of Pinealon have been marketed. Parenteral (subcutaneous, intramuscular, or intravenous) administration eliminates the bioavailability limitation but introduces the requirement for sterile formulation and injection.

5.2 Distribution

The proposed mechanism of action requires that Pinealon cross the blood-brain barrier and achieve intracellular and intranuclear concentrations sufficient for DNA interaction. The small molecular weight (418.41 g/mol) and moderate lipophilicity are consistent with potential blood-brain barrier penetration, though the charged nature of the molecule at physiological pH may limit passive diffusion. No blood-brain barrier penetration study (radiolabeled tracer, cerebrospinal fluid sampling, or imaging) has been published for Pinealon.

5.3 Metabolism and elimination

Tripeptides are rapidly metabolized by ubiquitous aminopeptidases, carboxypeptidases, and endopeptidases in plasma, liver, kidney, and other tissues. The plasma half-life of unmodified tripeptides is typically on the order of minutes, not hours. The metabolic products of Pinealon degradation would be the constituent free amino acids (glutamic acid, aspartic acid, arginine), which enter normal amino acid metabolism. No radiolabeled tracer study or mass spectrometric pharmacokinetic study has been published for Pinealon in any species.

5.4 Routes of administration

Three routes have been described in the published literature and in commercial formulations:

Oral: Capsule formulations (typically 200 micrograms to 10 milligrams per capsule) are the most widely available commercial presentation. The Khavinson group has used oral administration in the open-label clinical observations in elderly patients and in traumatic brain injury patients [13].

Sublingual: Sublingual ("lingual") formulations have been marketed for improved bioavailability. No comparative bioavailability study between oral and sublingual administration has been published.

Parenteral: Intraperitoneal administration has been used in rodent studies (typical doses 100 to 400 micrograms per kilogram body weight). Subcutaneous injection has been used in some research contexts. Intravenous administration has been described in animal models [10, 11, 12].

5.5 Dose ranges in published research

In vitro studies have typically used Pinealon at concentrations of 0.1 to 200 nanograms per milliliter (approximately 0.24 to 478 nanomolar) in cell culture medium [8, 11].

In vivo rodent studies have used doses of 100 to 400 micrograms per kilogram body weight administered intraperitoneally [10, 11, 12].

Clinical observations in humans have used oral doses of approximately 200 micrograms to 10 milligrams daily, with the capsule formulations typically providing 200 micrograms per capsule taken two to three times daily [13].

No formal dose-response study establishing a minimum effective dose, maximum tolerated dose, or optimal dosing regimen has been published for any route of administration in any species.

6. Preclinical Pharmacology

6.1 Antioxidant activity and cell viability

The foundational preclinical study of Pinealon's biological activity is the Khavinson et al. (2011) report published in *Rejuvenation Research*, which demonstrated that the synthetic tripeptide produces dose-dependent restriction of reactive oxygen species (ROS) accumulation in three cell types: rat cerebellar granule cells, human neutrophils, and rat pheochromocytoma (PC12) cells [8]. In cerebellar granule cells subjected to oxidative stress by hydrogen peroxide or glutamate excitotoxicity, Pinealon treatment reduced intracellular ROS levels and decreased necrotic cell death in a concentration-dependent manner. In neutrophils stimulated with phorbol myristate acetate (a receptor-independent ROS inducer) or with opsonized zymosan (a receptor-dependent inducer), Pinealon suppressed the respiratory burst and reduced extracellular ROS production. In PC12 cells, Pinealon reduced ROS accumulation and increased cell viability under oxidative stress conditions.

A notable observation from this study was that the antioxidant effect (suppression of ROS accumulation and reduction of cell death) saturated at relatively low Pinealon concentrations, while a separate cell cycle modulatory effect (stimulation of proliferative processes) continued at higher concentrations. This dissociation suggested that Pinealon produces at least two pharmacologically distinct effects: a direct or indirect antioxidant effect operative at low concentrations, and a gene-expression-mediated proliferative effect operative at higher concentrations, consistent with the proposed chromatin-interaction mechanism [8].

6.2 Neuroprotection in excitotoxicity and ischemia models

The Khavinson group has reported neuroprotective activity of Pinealon in several models of neuronal injury:

Homocysteine excitotoxicity: In rat cerebellar granule cells exposed to homocysteine (a sulfur-containing amino acid that acts as an NMDA receptor agonist and produces excitotoxic neuronal death at elevated concentrations), Pinealon treatment delayed ERK1/2 phosphorylation and reduced markers of apoptotic cell death [5].

Ischemia models: In rodent models of cerebral ischemia, Pinealon administration reduced caspase-3 expression, decreased p53 protein synthesis, and improved post-ischemic learning indices in the Morris water maze across both young and aged animals [5, 10].

Hypoxia models: In rats classified as hypoxia-sensitive by pre-screening behavioral paradigms, Pinealon administration increased SOD2 and GPX1 activity in brain tissue to levels comparable to those observed in hypoxia-resistant animals, normalizing the antioxidant enzyme deficit that correlates with hypoxia sensitivity [5, 8].

6.3 Dendritic spine preservation in Alzheimer's disease models

Khavinson, Linkova and colleagues (2017) reported that Pinealon prevents the loss of mushroom-shaped dendritic spines in primary cultures of hippocampal neurons from 5xFAD transgenic mice, a model of familial Alzheimer's disease that expresses five human mutations associated with early-onset amyloid pathology [11]. Mushroom spines are the morphological correlate of mature, stable synapses; their selective loss is a hallmark of amyloid-beta-induced synaptotoxicity and correlates with cognitive decline in Alzheimer's disease. At a concentration of 200 ng/mL, EDR peptide increased the number of mushroom spines by 71 percent under conditions of amyloid synaptotoxicity and returned this parameter to the normal (non-transgenic) level. A related tripeptide, KED (Lys-Glu-Asp), increased mushroom spines by 20 percent under the same conditions, indicating sequence-specific differences in potency [11].

In vivo confirmation was reported in 5xFAD-M mice (a variant crossed with the Thy1-YFP-M reporter line for dendritic spine visualization). Daily intraperitoneal administration of EDR peptide from 2 to 4 months of age tended to preserve dendritic spine number relative to untreated 5xFAD controls [19].

Molecular modeling studies supporting this work identified EDR binding sites in the promoter regions of CASP3, NES, GAP43, APOE, SOD2, PPARG, and GDX1, all of which encode proteins implicated in Alzheimer's disease pathogenesis [7].

6.4 Prenatal neuroprotection

Tchesnokova et al. (2012) reported that Pinealon protects rat offspring from prenatal hyperhomocysteinemia, a model in which maternal methionine loading during pregnancy produces elevated fetal homocysteine levels and postnatal cognitive deficits [12]. Offspring of methionine-loaded dams treated with Pinealon showed improved spatial orientation and learning ability in behavioral testing, alongside significant reductions in reactive oxygen species accumulation and necrotic cell counts in isolated cerebellar neurons, compared to untreated methionine-loaded controls. Notably, Pinealon did not reduce maternal or offspring homocysteine levels, indicating that the neuroprotective mechanism operates downstream of the homocysteine elevation rather than by reducing the toxic exposure itself [12].

6.5 Serotonin biosynthesis upregulation

Khavinson, Linkova and colleagues reported that Pinealon increases serotonin synthesis in neuronal cultures of rat cerebral cortex through epigenetic upregulation of tryptophan hydroxylase 1 (TPH1) expression [9, 16]. The proposed mechanism involves EDR peptide binding to the CCTGCC sequence in the TPH1 gene promoter, altering chromatin accessibility and facilitating TPH1 transcription. This activity positions Pinealon within the tryptophan-serotonin-melatonin biosynthetic cascade and provides a molecular rationale for reported effects on pineal function, circadian rhythm modulation, and sleep-wake cycle regulation, though direct in vivo demonstration of circadian or sleep effects through this mechanism has not been published [16].

6.6 Diabetes-related cognitive protection

Pinealon has been reported to maintain learning retention in rats with experimentally induced diabetes, suggesting neuroprotective activity in the context of diabetic encephalopathy [20]. The mechanism is proposed to involve the same antioxidant and anti-apoptotic pathways characterized in the ischemia and excitotoxicity models, given the well-established role of oxidative stress in diabetic neuronal damage.

7. Clinical Evidence Base

7.1 Traumatic brain injury

The most frequently cited clinical observation for Pinealon is an open-label study in 72 patients with traumatic brain injury, in which addition of Pinealon to standard rehabilitation therapy improved memory function in approximately 59.4 percent of patients, as assessed by standard neuropsychological testing [13]. The study was not randomized, was not blinded, did not include a concurrent control group receiving standard therapy alone (comparisons were to historical or baseline performance), and was published in a Russian-language journal. While the magnitude of the reported effect is clinically meaningful, the study design does not permit attribution of the improvement to Pinealon rather than to the natural recovery trajectory, placebo effect, or concomitant rehabilitation interventions.

7.2 Elderly cognitive decline

Pinealon has been administered orally to elderly patients with age-related cognitive decline in open-label clinical observations reported in Russian-language publications. Reported outcomes include improvement in short-term memory, environmental orientation, attention, and mental performance [13, 14]. These observations share the methodological limitations of the traumatic brain injury study: absence of randomization, absence of blinding, absence of concurrent control groups, and publication in journals with limited international peer review.

7.3 Cortexin clinical experience as context

Cortexin, the parent peptide mixture from which Pinealon was derived, has substantially greater clinical documentation. Multiple multicenter observational studies and controlled clinical investigations in ischemic stroke, traumatic brain injury, and pediatric neurological disorders have been published, predominantly in Russian neurology journals [14, 21]. While the Cortexin clinical record does not directly validate Pinealon (Cortexin contains many peptide components beyond EDR), it provides a broader clinical context suggesting that the peptide fraction

from which Pinealon was isolated has measurable neurological activity in human patients. The degree to which Pinealon itself accounts for Cortexin's clinical effects remains uncharacterized.

7.4 Absence of controlled clinical trials

No completed, peer-reviewed randomized controlled trial of Pinealon has been published in any language in journals indexed by PubMed, Embase, or the Cochrane Library as of the date of this monograph. No formal Phase 1 dose-escalation safety study, no Phase 2 proof-of-concept efficacy trial, and no Phase 3 registration trial has been conducted. No clinical trial registration for Pinealon appears in ClinicalTrials.gov, the EU Clinical Trials Register, or the WHO International Clinical Trials Registry Platform. The clinical evidence base therefore consists entirely of uncontrolled observational reports from a single research group and does not meet the evidentiary standards required for regulatory consideration in any major pharmaceutical jurisdiction.

8. Sourcing and Quality Verification

Research-grade Pinealon is supplied by multiple peptide synthesis companies and research chemical suppliers (Peptide Sciences, BenchChem, Smolecule, Spartan Peptides, BiotechPeptides, and others) at stated purities of 95 to 99 percent by HPLC. Supply is typically in milligram quantities (5 mg, 10 mg, 20 mg vials) as a lyophilized powder (trifluoroacetate or acetate salt). Pricing is moderate, in the range of \$30 to \$100 per 10 mg of research-grade material at the time of monograph preparation.

Capsule formulations marketed as dietary supplements are available through Russian and international supplement distributors, typically at 200 micrograms per capsule in capsule counts of 20 to 60. These formulations have not undergone pharmaceutical-grade quality review and should not be used for research applications where purity, identity, and dose accuracy are critical.

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

- Reversed-phase high-performance liquid chromatography area-percent purity (target: greater than 95 percent, ideally greater than 98 percent)
- Mass spectrometry confirmation of molecular weight (expected $[M+H]^+ = 419.19$ for the protonated species)
- Amino acid analysis to confirm the correct Glu:Asp:Arg ratio (1:1:1)
- Counterion identification (trifluoroacetate vs. acetate) if relevant to the experimental application
- Residual solvent analysis for solid-phase synthesis solvents (dimethylformamide, dichloromethane, acetonitrile, trifluoroacetic acid)
- Endotoxin testing (limulus amebocyte lysate assay) if the material will be used in cell-based or in vivo studies

Third-party analytical confirmation of vendor certificates of analysis is appropriate for any investigation where dose-response precision or mechanistic interpretation depends on compound identity and purity.

9. Reconstitution and Handling

Pinealon (as the lyophilized trifluoroacetate or acetate salt) is freely soluble in water at room temperature. For research applications, the compound is reconstituted in sterile water for injection, phosphate-buffered saline, or cell culture medium. Stock solutions are typically prepared at 1 to 10 mg/mL and stored in aliquots at minus 20 degrees Celsius or minus 80 degrees Celsius.

Reconstitution protocol: Add sterile water or buffer slowly along the wall of the vial to avoid foaming. Gently swirl (do not vortex) until fully dissolved. Filter through a 0.22 micrometer syringe filter for sterile applications. Aliquot into single-use volumes to avoid repeated freeze-thaw cycles.

Storage stability: The lyophilized powder is stable at minus 20 degrees Celsius for at least 24 months when stored desiccated and protected from light. Reconstituted aqueous solutions should be used within 7 days when stored at 2 to 8 degrees Celsius, or within 3 months when stored at minus 20 degrees Celsius in aliquots. Stability data for Pinealon in solution are limited; investigators should confirm peptide integrity by HPLC after storage if quantitative dose accuracy is important.

Handling precautions: Pinealon is not classified as hazardous by Globally Harmonized System criteria. Standard laboratory personal protective equipment (gloves, lab coat, eye protection) is appropriate. The compound is hygroscopic; the lyophilized powder should be equilibrated to room temperature before opening to prevent moisture absorption.

Compatibility: Pinealon is compatible with standard cell culture media (DMEM, RPMI, Neurobasal), phosphate-buffered saline, and normal saline. Compatibility with dimethyl sulfoxide has been reported for stock solution preparation at high concentrations, though the final DMSO concentration in biological assays should not exceed 0.1 percent to avoid solvent-mediated cytotoxicity. The compound should not be dissolved in strong acid or base, which would hydrolyze the peptide bonds.

10. Stack Interactions and Combinations

Formal drug-drug interaction studies have not been conducted for Pinealon in any species. The following considerations are based on the proposed mechanism of action, the known pharmacology of the constituent amino acids, and general principles of peptide pharmacology:

Serotonergic agents: The reported upregulation of tryptophan hydroxylase and serotonin synthesis by Pinealon raises a theoretical concern for additive serotonergic activity when combined with selective serotonin reuptake inhibitors (fluoxetine, sertraline, paroxetine, escitalopram), serotonin-norepinephrine reuptake inhibitors (venlafaxine, duloxetine), monoamine oxidase inhibitors, or tryptophan supplements. While the magnitude of the serotonin-stimulatory effect at achievable *in vivo* concentrations is likely small relative to the effects of pharmaceutical serotonergic agents, investigators should consider the theoretical risk of serotonin syndrome in concurrent administration.

Other neuroprotective peptides: Concurrent administration with other Khavinson bioregulatory peptides (Epithalon, KED, Cortexin) or with other neuroprotective peptides (Semax, Selank, Dihexa, BPC-157) has not been systematically studied. Additive or synergistic neuroprotective effects are theoretically plausible but uncharacterized. The Khavinson group has reported that EDR and KED produce qualitatively similar but quantitatively different effects on dendritic spine preservation, suggesting non-redundant mechanisms within the bioregulatory peptide class [11].

Antioxidant compounds: Concurrent administration with exogenous antioxidants (N-acetylcysteine, alpha-lipoic acid, vitamin C, vitamin E) may produce additive antioxidant effects. No interaction study has been conducted.

Cholinergic agents: No pharmacological interaction between Pinealon and acetylcholinesterase inhibitors (donepezil, rivastigmine, galantamine) has been characterized. The proposed mechanisms are non-overlapping (epigenetic gene

regulation vs. synaptic acetylcholine elevation), but both target neuroprotective endpoints, and concurrent administration in cognitive impairment research is theoretically feasible.

Melatonin: Given the proposed upregulation of the tryptophan-serotonin-melatonin biosynthetic pathway by Pinealon, concurrent exogenous melatonin administration would be expected to produce additive circadian and sleep-promoting effects. No interaction study has been conducted.

Enzymatic degradation interactions: Pinealon is likely degraded by ubiquitous aminopeptidases and carboxypeptidases. Co-administration with peptidase inhibitors (bestatin, amastatin) could theoretically increase Pinealon's bioavailability and extend its plasma half-life, though this interaction has not been studied.

11. Adverse Events and Safety Signal

11.1 Preclinical safety

No formal preclinical toxicology study (acute toxicity, subacute toxicity, chronic toxicity, genotoxicity, carcinogenicity, reproductive toxicity) has been published for Pinealon in peer-reviewed literature. The Khavinson group's published cell culture and rodent studies have not reported overt toxicity at the studied concentrations and doses, but these studies were designed as pharmacology investigations, not safety assessments, and did not include comprehensive toxicological endpoints.

The constituent amino acids (glutamic acid, aspartic acid, arginine) are all proteinogenic, endogenous, and generally recognized as safe as dietary components. The tripeptide itself, at the low doses used in published research (nanogram to microgram per milliliter in vitro; 100 to 400 micrograms per kilogram in vivo), is expected to present minimal acute toxicity risk based on first principles.

11.2 Clinical safety observations

The open-label clinical observations in traumatic brain injury patients and elderly patients with cognitive decline have not reported serious adverse events attributable to Pinealon [13]. However, the absence of reported adverse events in uncontrolled observations without systematic safety monitoring does not constitute evidence of safety. Spontaneous adverse event reporting in open-label observations is known to underestimate true adverse event rates relative to active surveillance in randomized trials.

11.3 Theoretical safety considerations

Serotonin excess: The reported upregulation of tryptophan hydroxylase and serotonin synthesis raises a theoretical concern for serotonin excess at high doses or in combination with serotonergic medications, though the magnitude of the effect at achievable concentrations is likely small.

Gene expression modulation: The proposed mechanism of direct chromatin interaction and gene expression modulation raises theoretical concerns about unintended gene activation or repression, particularly with chronic or high-dose administration. The proposed binding sites span both neuroprotective genes (SOD2, GPX1) and genes with complex regulatory roles (PPARG, APOE, CASP3). The consequences of chronic modulation of these genes in the context of normal physiology, as opposed to the pathological models in which the peptide has been studied, have not been characterized.

Immunogenicity: Tripeptides are generally too small to elicit adaptive immune responses. Anaphylaxis or hypersensitivity reactions to Pinealon have not been reported.

11.4 Pregnancy and lactation

No reproductive toxicology data have been published for Pinealon. The prenatal hyperhomocysteinemia study administered Pinealon to pregnant rats and reported improved offspring outcomes without overt maternal toxicity, but this single study does not constitute a reproductive toxicology assessment [12]. Use in pregnancy and lactation cannot be recommended or discouraged on the basis of the available evidence; formal reproductive toxicology studies would be required before any such recommendation.

12. Comparable Compounds Assessment

This section presents a structured comparative assessment of five neuroprotective peptide compounds against Pinealon, 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 peptide-class neuroprotective agents with substantive preclinical or clinical pharmacology relevant to cognitive protection, neuronal survival, or bioregulatory gene modulation.

12.1 Semax (Met-Glu-His-Phe-Pro-Gly-Pro)

Mechanism: Synthetic analog of the ACTH(4-10) fragment. Nootropic and neuroprotective activity mediated through modulation of brain-derived neurotrophic factor (BDNF), nerve growth factor (NGF), and TrkB receptor signaling. No direct chromatin interaction proposed; mechanism is receptor-mediated and neurotrophic.

Development trajectory: Developed at the Institute of Molecular Genetics, Russian Academy of Sciences. Registered in the Russian Federation for the treatment of ischemic stroke, dyscirculatory encephalopathy, optic nerve atrophy, and cognitive impairment. Multiple open-label and controlled clinical studies have been published, including a randomized controlled trial in acute ischemic stroke patients demonstrating improved neurological outcomes [22, 23].

Novelty: Moderate. The ACTH-fragment nootropic mechanism is well established. Semax represents an optimized variant with improved metabolic stability.

Effect size: Moderate to substantial in published stroke and cognitive studies, though many are from Russian clinical settings with limited independent replication.

Promising potential: Moderate. The compound is registered as a pharmaceutical in Russia and Ukraine with clinical use experience spanning several decades, but has not been advanced to regulatory submission in ICH-recognized jurisdictions.

Side-effect profile: Reported as generally well tolerated in published studies. No serious adverse events attributable to the compound have been identified in published literature. The peptide nature and the low doses used minimize systemic toxicity risk.

Overall validation: Moderate. Substantially more clinical evidence than Pinealon, including at least one published randomized controlled trial. Registered as a medicine in two jurisdictions. Limited independent Western replication.

Comparison to Pinealon: Semax has a substantially more developed clinical evidence base, a more conventional receptor-mediated mechanism, and registered pharmaceutical status. Pinealon has a more novel (though less validated) proposed mechanism of action and a different target gene profile. Semax is the more clinically validated compound; Pinealon is the more mechanistically unconventional one.

12.2 Selank (Thr-Lys-Pro-Arg-Pro-Gly-Pro)

Mechanism: Synthetic analog of the endogenous tetrapeptide tuftsin (Thr-Lys-Pro-Arg) with a Pro-Gly-Pro tripeptide stabilization fragment. Anxiolytic and nootropic activity mediated through modulation of monoamine neurotransmitter metabolism, BDNF expression, and enkephalin degradation. Registered in the Russian Federation as an anxiolytic.

Development trajectory: Developed at the Institute of Molecular Genetics, Russian Academy of Sciences (the same institute as Semax). Registered in the Russian Federation for anxiety disorders. Published preclinical and clinical studies include anxiolytic, nootropic, and immunomodulatory endpoints [24].

Novelty: Moderate. The tuftsin-derived anxiolytic mechanism is distinct from conventional benzodiazepine or SSRI anxiolysis.

Effect size: Moderate in published anxiety and cognitive studies. Anxiolytic effect reported as comparable to benzodiazepines without sedation or dependence.

Promising potential: Moderate. Registered as a pharmaceutical in Russia. Not advanced to ICH-jurisdiction regulatory submission.

Side-effect profile: Reported as well tolerated with minimal adverse events. No sedation, no dependence, no withdrawal reported.

Overall validation: Moderate. More clinical evidence than Pinealon. Registered pharmaceutical status in one jurisdiction.

Comparison to Pinealon: Selank has a more established clinical profile and a more conventional mechanism. Pinealon has a broader proposed gene expression modulation profile but less clinical validation. The two compounds target different therapeutic endpoints (Selank: anxiety and nootropic; Pinealon: neuroprotection and geroprotection) and are not directly competitive.

12.3 Epithalon (Ala-Glu-Asp-Gly)

Mechanism: Synthetic tetrapeptide developed by the Khavinson group as a pineal gland bioregulator. Proposed mechanism involves activation of telomerase reverse transcriptase (hTERT) gene expression, resulting in telomere elongation, and up-regulation of melatonin synthesis by the pineal gland. The geroprotective hypothesis is that Epithalon counteracts age-related pineal gland involution and telomere shortening [25].

Development trajectory: Developed at the Saint Petersburg Institute of Bioregulation and Gerontology (the same institute as Pinealon). Preclinical studies include telomerase activation in human somatic cells, lifespan extension in rodent models, and melatonin secretion restoration in aged primates. No formal clinical trials have been published [25, 26].

Novelty: High. The telomerase-activation-by-short-peptide mechanism is highly unconventional and, if validated, would represent a fundamentally new approach to cellular aging.

Effect size: Substantial in the Khavinson group's rodent lifespan studies (reported 20 to 30 percent lifespan extension), but these studies have not been independently replicated with modern biological aging endpoints.

Promising potential: Low to moderate. No clinical trials, no regulatory submission. Strong theoretical interest but dependent on independent validation.

Side-effect profile: No reported adverse events in preclinical studies. Theoretical concern about telomerase activation and oncogenesis has not been addressed by long-term safety studies.

Overall validation: Low. All evidence from a single research group without independent replication. Mechanistic claims are extraordinary and require extraordinary evidence.

Comparison to Pinealon: Epithalon and Pinealon originate from the same laboratory and share the same proposed class mechanism (direct chromatin interaction by ultrashort peptides). They differ in target organ (pineal gland/telomere vs. cerebral cortex/neuroprotection) and in proposed gene targets (hTERT vs. TPH1/SOD2/CASP3). Neither compound has clinical trial data. The validation challenges are identical: single-group evidence base, absence of independent replication, unconventional mechanism requiring structural biology confirmation.

12.4 Cortexin (complex peptide fraction)

Mechanism: Complex mixture of low-molecular-weight peptides (molecular weight less than 10,000 Da) extracted from bovine cerebral cortex by acid hydrolysis and ultrafiltration. Cortexin is the first-generation cytomaxe from which Pinealon was derived. Mechanism is proposed as multi-target neuroprotection through the combined activity of multiple peptide components acting on antioxidant, anti-apoptotic, neurotrophic, and anti-inflammatory pathways [14].

Development trajectory: Developed at the Military Medical Academy in Leningrad. Registered in the Russian Federation for ischemic stroke, traumatic brain injury, viral encephalitis, epilepsy, and cognitive impairment. Extensive clinical use in Russian and post-Soviet clinical practice spanning over 30 years. Multiple published clinical studies, including multicenter observational studies in ischemic stroke patients [14, 21].

Novelty: Low. Complex organ-derived peptide fractions represent an older pharmacological paradigm. The novelty of Cortexin lies in its clinical track record rather than its mechanism.

Effect size: Moderate in published stroke and traumatic brain injury studies. Reported improvements in neurological outcomes, cognitive function, and rehabilitation parameters.

Promising potential: Moderate within the Russian/post-Soviet pharmaceutical framework. Not advanced to ICH-jurisdiction regulatory submission.

Side-effect profile: Reported as well tolerated across decades of clinical use. No serious adverse events identified in published literature.

Overall validation: Moderate to high within the Russian evidence framework. Registered pharmaceutical with extensive clinical use. Limited international peer review.

Comparison to Pinealon: Cortexin is the parent mixture from which Pinealon was derived and has substantially greater clinical validation (registered indication, multicenter studies, decades of clinical experience). Pinealon is the defined-sequence, single-entity derivative with a more precisely characterized (though less validated) mechanism. The relationship is analogous to a natural product extract versus its isolated active constituent. Cortexin is the more clinically validated compound; Pinealon is the more mechanistically defined one.

12.5 Dihexa (N-hexanoyl-Tyr-Ile-[6]-aminohexanoic acid)

Mechanism: Synthetic modified dipeptide analog of angiotensin IV. Potent activator of hepatocyte growth factor (HGF) / c-Met receptor signaling. Procognitive and neurotrophic activity demonstrated in rodent models at picomolar concentrations. Orally bioavailable. Proposed as a disease-modifying agent for Alzheimer's disease through promotion of dendritic spine formation and synaptic connectivity via the HGF/c-Met pathway [27].

Development trajectory: Developed at Washington State University by Joseph Harding and colleagues. Advanced through extensive preclinical evaluation including scopolamine-induced cognitive deficit models, aged rat cognitive models, and dendritic spine formation assays. No clinical trials have been published. The compound has been widely adopted in the research peptide community [27, 28].

Novelty: High. The HGF/c-Met procognitive mechanism is distinct from cholinergic, serotonergic, or glutamatergic approaches. The potency (active at picomolar concentrations) is exceptional.

Effect size: Substantial in preclinical models. Reported procognitive effects at 10,000-fold lower doses than conventional peptide nootropics.

Promising potential: Moderate. Extensive preclinical data from a reputable Western academic laboratory. No clinical translation to date. Oral bioavailability is a significant practical advantage.

Side-effect profile: No reported adverse events in preclinical studies. The HGF/c-Met pathway has oncogenic potential; long-term safety implications are uncharacterized.

Overall validation: Moderate. Strong preclinical evidence base from an independent Western laboratory. No clinical data.

Comparison to Pinealon: Dihexa and Pinealon both target neuroprotection and cognitive enhancement through peptide-based approaches but differ fundamentally in mechanism (HGF/c-Met receptor activation vs. proposed chromatin interaction), potency (picomolar vs. nanomolar effective concentrations), and evidence base (independent Western academic validation vs. single-group Russian validation). Dihexa has substantially more credible preclinical validation from an independent laboratory; Pinealon has a broader proposed gene expression modulation profile. Neither has clinical trial data.

12.6 Comparator summary

The five comparators span the spectrum of neuroprotective peptide development. **Semax** and **Selank** represent the most clinically validated compounds, with registered pharmaceutical status and published controlled studies, though principally within the Russian regulatory and academic framework. **Cortexin** is the parent mixture of Pinealon with extensive clinical use but represents an older pharmacological paradigm of complex peptide fractions. **Epithalon** shares Pinealon's origin laboratory, proposed class mechanism, and evidence limitations (single-group data, absence of independent replication). **Dihexa** represents the most mechanistically credible preclinical neuroprotective peptide candidate from an independent Western laboratory but lacks clinical data.

Pinealon occupies a distinctive but scientifically precarious position in this landscape. Its proposed mechanism (direct chromatin interaction and epigenetic gene regulation by a tripeptide) is the most novel claim among the comparators and, if validated, would represent a fundamentally new pharmacological paradigm. However, the evidence supporting this mechanism derives entirely from a single research group, has not been independently replicated, and has not been validated by the structural biology methods (crystallography, cryo-EM, ChIP-seq) that would be required to establish the proposed binding geometry and genomic target sites

with confidence. The clinical evidence base (open-label observations without randomization or blinding) does not meet contemporary evidentiary standards for efficacy claims.

The principal question for the research positioning of Pinealon is whether the proposed chromatin-interaction mechanism is genuine or artifactual. If genuine, the compound represents a potentially transformative approach to tissue-specific gene regulation with implications extending well beyond neuroprotection. If the observed biological activity is real but mediated by a different mechanism (for example, antioxidant activity of the constituent amino acids, non-specific peptide-membrane interactions, or conventional signaling through unidentified receptors), the compound retains interest as a neuroprotective agent but loses the mechanistic novelty that is its principal differentiating feature. Independent replication of the core mechanistic claims is the critical next step for the field.

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This monograph is provided for research use only. Not for medical, diagnostic, or therapeutic purposes. Not for human consumption. The evidence base for Pinealon consists predominantly of publications from a single research group without independent replication by Western academic laboratories. The proposed chromatin-interaction mechanism has not been validated by independent structural biology methods. No randomized controlled trial, no formal safety study, and no regulatory approval exists for this compound in any ICH-recognized jurisdiction. Investigators should weight the evidence accordingly and should obtain analytical confirmation of identity and purity on every lot of research-grade material.