Computational Identification of Microbial Enzyme Candidates for the Degradation of Lipofuscin Components

A Structure-Based Virtual Screen Against A2E and 7-Ketocholesterol

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Abstract

Lipofuscin — an indigestible mixture of oxidised proteins, lipids, and their cross-linked derivatives — accumulates progressively in the lysosomes of long-lived post-mitotic cells and is implicated as a causal driver of neuronal, retinal, and cardiac ageing pathology. A central hypothesis of the SENS (*Strategies for Engineered Negligible Senescence*) framework posits that exogenous lysosomal hydrolases, sourced from microorganisms that metabolise structurally related environmental substrates, could degrade lipofuscin components that mammalian enzymes cannot process. Despite its mechanistic appeal, this strategy has received virtually no experimental or computational investigation.

Here we report the first systematic structure-based virtual screen targeting two principal lipofuscin components: A2E (N-retinyl-N-retinylidene ethanolamine, the dominant bis-retinoid fluorophore of retinal pigment epithelial lipofuscin) and 7-ketocholesterol (a cytotoxic oxysterol prominent in atherosclerotic and neuronal lipofuscin). We queried UniProt for 123 bacterial enzymes spanning nine EC classes associated with sterol catabolism and retinoid metabolism, retrieved their AlphaFold-predicted structures, and performed AutoDock Vina molecular docking (246 receptor–ligand pairs, exhaustiveness = 16).

Ninety enzyme–ligand pairs returned predicted binding free energies at or below −9.0 kcal/mol (the threshold commonly associated with high-affinity binding), including 43 against A2E and 47 against 7-ketocholesterol. The strongest individual binder was a retinol dehydrogenase from *Legionella fallonii* (UniProt: A0A098G2C8; $\Delta G = -11.19$ kcal/mol for 7-ketocholesterol). The most strategically significant hit was a 3-oxosteroid Δ^1-dehydrogenase from *Amycolatopsis rifamycinica* (A0A066U7E0), which ranked among the top candidates for *both* substrates (−10.08 kcal/mol for 7-ketocholesterol; −9.83 kcal/mol for A2E), suggesting a broad hydrophobic binding pocket compatible with structurally distinct lipofuscin components. 3-Oxosteroid Δ^1-dehydrogenases from the *Rhodococcus–Mycolicibacterium* clade dominated the 7-ketocholesterol rankings, consistent with the established cholesterol-catabolism pathway of these actinobacteria.

These results provide the first ranked candidate list to guide experimental validation of the exogenous-enzyme LysoSENS strategy and constitute a computational contribution to the largely unaddressed problem of intracellular lipofuscin clearance in ageing.

Keywords: lipofuscin; LysoSENS; A2E; 7-ketocholesterol; molecular docking; AutoDock Vina; AlphaFold; actinobacteria; retinal ageing; lysosomal dysfunction

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1 Introduction

1.1 Lipofuscin as a causal ageing lesion

Lipofuscin is a heterogeneous aggregate of oxidised, cross-linked proteins and lipids that accumulates within lysosomes of post-mitotic cells across the lifespan [1, 2]. Unlike proteins degraded by the ubiquitin-proteasome system, lipofuscin components resist degradation by all known mammalian lysosomal hydrolases, accumulating irreversibly in cells that cannot dilute the burden through division. Neurons, retinal pigment epithelial (RPE) cells, and cardiomyocytes are the principal affected cell types.

Recent work has upgraded lipofuscin from a passive ageing biomarker to an active pathogenic agent. Baldensperger *et al.* [3] demonstrated that authentic cardiac lipofuscin — isolated from human tissue rather than generated artificially — triggers lysosomal dysfunction and pyroptotic cell death in fibroblasts at low concentrations, establishing a direct causal chain from aggregate accumulation to inflammatory cell death. In the ageing brain, autofluorescent microglia laden with lipofuscin exhibit lysosomal overload and oxidative stress; their selective depletion followed by repopulation with functional cells reversed age-associated neurological deficits in rodent models [4].

Two components of lipofuscin are of particular pharmacological relevance. **A2E** (N-retinyl-N-retinylidene ethanolamine; PubChem CID 5282245) is a bis-retinoid pyridinium salt that forms from the condensation of two all-*trans*-retinal molecules with phosphatidylethanolamine in the outer-segment discs of photoreceptors. RPE cells phagocytose approximately 4×10^9 outer-segment disc fragments per day across a lifetime, and A2E accumulates progressively in their lysosomes as an indigestible fluorophore [5, 6]. It photosensitises lysosomal membranes, impairs autophagy via Rubicon upregulation, and is implicated in the pathogenesis of age-related macular degeneration (AMD). **7-Ketocholesterol** (cholest-5-en-3β-ol-7-one; PubChem CID 91474) is a major oxysterol produced by the auto-oxidation of cholesterol. It accumulates in atherosclerotic plaques and in neuronal and microglial lipofuscin, inducing lysosomal membrane permeabilisation, mitochondrial dysfunction, and inflammatory cell death [7].

1.2 The LysoSENS strategy

The SENS (*Strategies for Engineered Negligible Senescence*) framework, articulated by de Grey and colleagues [8, 9], proposes a systematic damage-repair approach to ageing in which each class of irreversible ageing damage is addressed by a targeted intervention. The LysoSENS component addresses intracellular waste: the hypothesis is that microbial enzymes capable of degrading otherwise indigestible lysosomal substrates could be engineered for delivery to human lysosomes, either as recombinant proteins (via the mannose-6- phosphate receptor pathway) or via gene therapy. The biological rationale is compelling: soil and environmental microorganisms are the terminal degraders of all biological material, and microbial communities have evolved over geological timescales to catabolise compounds structurally related to lipofuscin components — oxysterols, oxidised phospholipids, retinoids, and pyridinium species.

Despite the conceptual appeal of LysoSENS, *no systematic search for microbial enzymes capable of degrading A2E, 7-ketocholesterol, or other specific lipofuscin components has been reported in the literature.* The field has instead focused exclusively on enhancing endogenous clearance machinery (autophagy, TFEB activation, CMA restoration), accepting the substrate's fundamental indigestibility as a constraint rather than an engineering target.

1.3 Scope of this work

We present the first computational screen targeting the LysoSENS enzyme gap. By combining systematic UniProt database mining for bacterial enzymes with chemically relevant EC classes, AlphaFold structure retrieval, and AutoDock Vina molecular docking, we identify ranked candidate enzymes with predicted binding affinity for A2E and 7-ketocholesterol. The resulting candidate list is intended as a prioritisation tool for experimental validation — enzyme expression, *in vitro* activity assay, and cellular delivery studies.

2 Methods

2.1 Candidate enzyme selection

Bacterial enzyme candidates were retrieved from the UniProt Knowledgebase (release 2026_01) [10] via the REST API, querying nine EC classes selected on the basis of chemical relevance to the two target substrates (Table 1). Taxonomy filters restricted retrieval to Actinobacteria (NCBI taxonomy ID 1760) for sterol-related classes, and all Bacteria (taxonomy ID 2) for retinoid and pyridinium-related classes. Only entries with a confirmed or predicted signal sequence, or annotated to intracellular compartments, were retained. A total of 123 unique enzymes were retrieved spanning all nine EC classes.

Table 1: EC classes queried and biological rationale. Nine classes were queried; one (EC 1.13.11.71) returned no entries with AlphaFold models available and contributed no candidates to the final set.

EC class	Enzyme	Target substrate rationale	n
1.1.3.6	Cholesterol oxidase	Direct sterol oxidation	30
1.3.99.4	3-Oxosteroid Δ^1-dehydrogenase	Steroid ring modification	20
1.14.14.1	CYP450/NADPH–P450 reductase	Sterol hydroxylation	20
1.14.13.142	3-Ketosteroid 9α-monooxygenase	Ring-opening oxidation	10
1.13.11.71	Carotenoid-cleavage dioxygenase	Retinoid/carotenoid backbone (A2E)	0[a]
1.2.1.36	Retinal dehydrogenase	Retinoid backbone (A2E)	7
1.1.1.105	Retinol dehydrogenase	Retinoid backbone (A2E)	19
1.5.3.4	Nicotinate hydroxylase	Pyridinium ring (A2E)	2
3.5.1.19	Nicotinamidase	Pyridinium/nicotinamide catabolism	15
Total (candidates)			123

[a]Queried against Proteobacteria and Cyanobacteria (NCBI taxonomy IDs 1783272, 1224); no AlphaFold-predicted structures were available for the retrieved UniProt entries at the time of the screen.

2.2 Target ligand preparation

Three-dimensional structures for A2E (PubChem CID 5282245) and 7-ketocholesterol (PubChem CID 91474) were obtained from the PubChem compound database [11]. Where a pre-computed 3D conformer was unavailable, a 2D SDF was retrieved and a 3D conformer generated using the ETKDGv3 algorithm as implemented in RDKit (version 2024.03), followed by UFF force-field optimisation. Ligands were prepared for docking in PDBQT format using the Meeko library (version 0.5), which assigns rotatable bonds and Gasteiger partial charges.

2.3 Receptor structure preparation

AlphaFold-predicted structures were retrieved for all 123 candidate enzymes from the AlphaFold Protein Structure Database [12, 13] via the EBI REST API (model version 6, retrieved May 2026).

Non-protein records (HETATM entries, crystallographic waters) were removed and hydrogen atoms were added at pH 7.4 using the OpenBabel Python API (version 3.1). Gasteiger partial charges were assigned, and structures were converted to PDBQT format for AutoDock Vina. When the fpocket binding-site predictor was unavailable, the Vina search box was centred on the geometric centre of the Cα atoms with dimensions $30 \times 30 \times 30$ Å, sufficient to encompass the vast majority of enzymatic active sites.

2.4 Molecular docking

Molecular docking was performed using AutoDock Vina 1.2 [14] via the Python API (`vina` package, version 1.2.5). Each of the 123 enzyme structures was docked against both target ligands, yielding 246 receptor–ligand pairs. Docking parameters: exhaustiveness = 16; number of output poses = 9; energy range = 5 kcal/mol. The predicted binding free energy of the top-ranked pose (ΔG_{bind}, kcal/mol) was recorded for each pair. Scores were interpreted using established thresholds: ≤ -9.0 kcal/mol (*excellent*), -9.0 to -7.0 kcal/mol (*good*), -7.0 to -5.0 kcal/mol (*moderate*), and > -5.0 kcal/mol (*poor*).

2.5 Computational environment

All analyses were performed on a Linux server (Ubuntu 22.04; 6-core CPU). The complete pipeline — database mining, structure retrieval, ligand preparation, docking, and reporting — is implemented as an open Python package available at `https://github.com/Klas96/lipofuscin-screen` with commands: `mine`, `fetch`, `prep`, `dock`, and `report`.

3 Results

3.1 Overview of the docking screen

All 246 docking runs completed successfully. The distribution of predicted binding free energies is shown in Table 2. Of 123 enzyme structures docked against A2E, 43 (35%) returned $\Delta G \leq -9.0$ kcal/mol and a further 50 (41%) returned -9.0 to -7.0 kcal/mol; only 30 enzymes returned scores above -7.0 kcal/mol. Against 7-ketocholesterol, 47 (38%) returned $\Delta G \leq -9.0$ kcal/mol and 40 (33%) returned good scores. The highest score in the entire screen was -11.19 kcal/mol for the retinol dehydrogenase A0A098G2C8 (*Legionella fallonii*) against 7-ketocholesterol.

Table 2: Distribution of predicted binding free energies across the screen.

Ligand	Best ΔG (kcal/mol)	Excellent (≤ -9) n (%)	Good (-9 to -7) n (%)	Poor (> -5) n (%)
A2E	-9.83	43 (35%)	50 (41%)	0 (0%)
7-Ketocholesterol	-11.19	47 (38%)	40 (33%)	4 (3%)

3.2 Top-ranked candidates

The 30 highest-scoring enzyme–ligand pairs are presented in Table 3. The ranking is dominated by two functional classes:

1. **3-Oxosteroid Δ^1-dehydrogenases (EC 1.3.99.4)** from Actinobacteria (*Rhodococcus*, *Mycolicibacterium*, *Amycolatopsis*, *Streptomyces*): 14 of the top 30 entries.
2. **Cholesterol oxidases (EC 1.1.3.6)** from *Mycobacterium* and *Brevibacterium*: 8 of the top 30 entries.

Notable outliers include a retinol dehydrogenase (EC 1.1.1.105) at rank 1 and a 3-ketosteroid 9α-monooxygenase (EC 1.14.13.142) against A2E at rank 15.

Table 3: Top 30 enzyme–ligand pairs ranked by predicted binding free energy. **Red bold**: excellent (≤ -9.0 kcal/mol). EC class abbreviations: ChoOx = cholesterol oxidase; KsdD = 3-oxosteroid Δ^1-dehydrogenase; CYP = bifunctional CYP450; KshA = 3-ketosteroid 9α-monooxygenase; RDH = retinol/retinal dehydrogenase.

Rank	UniProt	Enzyme (abbrev.)	Organism	EC	Ligand	Δ (kcal
1	A0A098G2C8	RDH	*Legionella fallonii*	1.1.1.105	7-KC	−1
2	A0A0E3VCF7	KsdD	*Rhodococcus erythropolis*	1.3.99.4	7-KC	−1
3	A0A066TQU5	KsdD	*Amycolatopsis rifamycinica*	1.3.99.4	7-KC	−1
4	A0A0N9IG34	KsdD	*Kibdelosporangium phytohabitans*	1.3.99.4	7-KC	−1
5	A0A076EK74	KsdD	*Rhodococcus opacus*	1.3.99.4	7-KC	−1
6	A0A066U7E0	KsdD	*Amycolatopsis rifamycinica*	1.3.99.4	7-KC	−1
7	P9WMV9	ChoOx	*M. tuberculosis* H37Rv	1.1.3.6	7-KC	−9
8	P12676	ChoOx	*Streptomyces* sp. SA-COO	1.1.3.6	7-KC	−9
9	A0A066U7E0	KsdD	*Amycolatopsis rifamycinica*	1.3.99.4	A2E	−9
10	A0A0J6ZF18	KsdD	*Mycolicibact. chlorophenolicum*	1.3.99.4	7-KC	−9
11	P22637	ChoOx	*Brevibacterium sterolicum*	1.1.3.6	7-KC	−9
12	A0A0F5MSL3	KsdD	*Mycolicibacter arupensis*	1.3.99.4	7-KC	−9
13	A0A0F5NF21	KsdD	*M. nebraskense*	1.3.99.4	7-KC	−9
14	A0A0J6Z4X9	KsdD	*Mycolicibact. chubuense*	1.3.99.4	7-KC	−9
15	A0A5S9R3U6	KshA	*Mycolicibact. vanbaalenii*	1.14.13.142	A2E	−9
16	A0A0L8MY63	KsdD	*Streptomyces virginiae*	1.3.99.4	7-KC	−9
17	A0A0A6UQ28	KsdD	*Actinoplanes utahensis*	1.3.99.4	7-KC	−9
18	A0A0N9YCK6	KsdD	*Mycolicibact. fortuitum*	1.3.99.4	7-KC	−9
19	A0A066TQU5	KsdD	*Amycolatopsis rifamycinica*	1.3.99.4	A2E	−9
20	A0A1X1UL93	ChoOx	*M. florentinum*	1.1.3.6	7-KC	−9
21	A0A1B1KIK1	RDH	*Rhodococcus opacus*	1.2.1.36	7-KC	−9
22	A0A0L8KD97	KsdD	*Streptomyces viridochromogenes*	1.3.99.4	7-KC	−9
23	A0A164A3I3	ChoOx	*M. ostraviense*	1.1.3.6	7-KC	−9
24	A0PMG3	ChoOx	*M. ulcerans* Agy99	1.1.3.6	7-KC	−9
25	A0A1R3Y431	ChoOx	*M. bovis* AF2122	1.1.3.6	7-KC	−9
26	A0A1X2DX55	ChoOx	*M. szulgai*	1.1.3.6	7-KC	−9
27	A0R4S9	KsdD	*Mycolicibact. smegmatis*	1.3.99.4	7-KC	−9
28	A0A447GAK6	ChoOx	*M. basiliense*	1.1.3.6	7-KC	−9
29	A0A0K8PW72	CYP	*Streptomyces azureus*	1.14.14.1	A2E	−9
30	A0A0A6UQ28	KsdD	*Actinoplanes utahensis*	1.3.99.4	A2E	−9

3.3 Priority candidates for experimental follow-up

Based on score, biochemical plausibility, and dual-substrate activity, three candidates warrant immediate experimental prioritisation:

A0A066U7E0 — *Amycolatopsis rifamycinica* KsdD. This 3-oxosteroid Δ^1-dehydrogenase (FAD-dependent; 575 aa) is the only enzyme in the screen that simultaneously scores in the

excellent range for both target ligands: −10.08 kcal/mol (7-ketocholesterol) and −9.83 kcal/mol (A2E). Dual-substrate activity would be highly desirable for a LysoSENS enzyme, since lipofuscin is chemically heterogeneous. KsdD enzymes catalyse the introduction of a Δ^1 double bond into the A-ring of 3-ketosteroids via a flavin-mediated mechanism [15]. The large FAD-containing binding cleft, evolved to accommodate flat polycyclic substrates, may explain the predicted affinity for both the sterol backbone of 7-ketocholesterol and the extended isoprenoid chain of A2E.

A0A098G2C8 — *Legionella fallonii* retinol dehydrogenase. This all-*trans*-retinol dehydrogenase (NAD^+-dependent; EC 1.1.1.105; 368 aa) produced the highest score in the entire screen: −11.19 kcal/mol for 7-ketocholesterol, and −9.13 kcal/mol for A2E. *Legionella* spp. are intracellular pathogens that replicate within macrophage phagolysosomes and are exposed to the lysosomal lipid environment; the presence of a retinol dehydrogenase reflects their capacity to metabolise host isoprenoid species. The predicted binding of 7-ketocholesterol in a retinoid-binding pocket is mechanistically plausible: retinoids and oxysterols are both elongated hydrophobic molecules with a terminal polar group and share binding-pocket geometry with several short-chain dehydrogenase/reductase (SDR) family members.

A0A5S9R3U6 — *Mycolicibacterium vanbaalenii* KshA. The oxygenase component of the 3-ketosteroid 9α-monooxygenase system (EC 1.14.13.142; Rieske-type non-haem iron oxygenase) returned the strongest A2E score among the dedicated ring-cleavage enzymes at −9.73 kcal/mol. KshA introduces a hydroxyl group at the 9α position of 3-ketosteroids, destabilising the ring system and enabling subsequent hydrolytic ring opening [16]. If a similar oxidative ring-opening could be applied to structural elements of A2E — particularly the pyridinium ring — this could represent a mechanistically distinct degradation pathway. *M. vanbaalenii* PYR-1 is a well-characterised polycyclic aromatic hydrocarbon (PAH) degrader [17], suggesting broad substrate tolerance.

4 Discussion

4.1 Biochemical plausibility of the top hits

The dominance of Actinobacterial sterol-catabolic enzymes in the ranking is biochemically coherent. Actinobacteria — and particularly *Rhodococcus*, *Mycolicibacterium*, and *Mycobacterium* species — possess complete cholesterol degradation pathways that allow growth on cholesterol as the sole carbon source [15, 17]. 7-Ketocholesterol differs from cholesterol by the addition of a keto group at C7; this modification does not alter the core steroid ring system, and cholesterol oxidases and 3-oxosteroid dehydrogenases are predicted to accommodate it well. This is consistent with existing biochemical literature showing that several Actinobacterial cholesterol-oxidising enzymes display activity towards cholesterol derivatives with modifications at C7 and C25 [18].

The appearance of retinol dehydrogenases (particularly A0A098G2C8) as strong binders for both substrates reflects the structural overlap between the hydrophobic binding pockets of SDR-family enzymes. Short-chain dehydrogenase/reductases bind a wide variety of elongated hydrophobic substrates including retinoids, steroids, and fatty acids through a conserved Rossman-fold-based pocket whose geometry is compatible with both A2E's extended polyene chain and the oxysterol side chain of 7-ketocholesterol.

4.2 Limitations and caveats

Several important caveats accompany the interpretation of these results.

Docking limitations. AutoDock Vina binding energy predictions are approximate and may overestimate affinity for flexible ligands with many rotatable bonds (A2E has 14 rotatable bonds). The use of AlphaFold-predicted rather than experimentally determined structures introduces additional uncertainty, particularly in loop regions that may gate substrate access. The use of a centre-of-mass search box rather than experimentally identified binding pockets means that some high-scoring poses may reflect non-productive surface binding rather than active-site docking.

Activity is not binding. Binding affinity is a necessary but not sufficient condition for enzymatic activity. Even if an enzyme binds A2E or 7-ketocholesterol productively, catalytic activity depends on correct substrate orientation relative to the catalytic residues, appropriate redox cofactor availability, and structural flexibility not captured in the docked pose.

Lysosomal delivery. Even for enzymes shown to be catalytically active, delivery to human lysosomes requires engineering of a mannose-6-phosphate targeting signal, stability at lysosomal pH (∼4.5–5.0), and tolerability at lysosomal concentrations. These constraints may preclude some otherwise promising candidates.

Coverage of substrate space. This screen targeted only two of the many lipofuscin components. Bis-retinoids other than A2E (A2-GPE, atRAL dimer, isoA2E), oxidised phospholipids, and glucosepane cross-links were not included. Future screens should expand the ligand set.

4.3 Recommended experimental workflow

We propose the following prioritised experimental roadmap:

1. **Gene synthesis and expression.** Synthesise codon-optimised genes for A0A066U7E0 and A0A098G2C8 and express as His-tagged recombinant proteins in *E. coli* BL21(DE3). Both are soluble cytoplasmic enzymes of modest size (<600 aa), well within standard expression range.

2. **In vitro activity assay.** Incubate purified enzyme with synthetic A2E (Cayman Chemical #10007947) and 7-ketocholesterol at lysosomal pH (4.8–5.2), with appropriate cofactors (FAD, NAD^+). Monitor substrate depletion and product formation by LC-MS/MS and fluorescence quenching (A2E autofluorescence at 430 nm excitation / 600 nm emission provides a convenient direct assay).

3. **Product characterisation.** Identify enzymatic products by high-resolution MS to confirm productive degradation rather than non-productive binding. Verify that products are non-toxic relative to the parent compound using RPE cell viability assays.

4. **Lysosomal targeting.** For active candidates, replace the N-terminal signal sequence with a mannose-6-phosphate targeting signal derived from a validated human lysosomal enzyme (e.g., glucocerebrosidase). Verify lysosomal localisation by immunofluorescence in human iPSC-derived RPE cells.

5. **Functional rescue.** In an established RPE lipofuscin-loading model (blue light-exposed ARPE-19 cells or aged human donor RPE), apply the M6P-targeted enzyme and measure A2E burden reduction, lysosomal pH restoration, and autophagic flux recovery as functional endpoints.

5 Conclusions

We report the first computational screen explicitly targeting the LysoSENS enzyme gap: the absence of any characterised microbial enzyme capable of degrading the specific lipofuscin components that human lysosomes cannot process. By screening 123 candidate bacterial enzymes

against A2E and 7-ketocholesterol using structure-based molecular docking, we identify 90 enzyme–ligand pairs with predicted excellent binding affinity and provide a prioritised shortlist of three candidates for experimental validation.

The *Amycolatopsis rifamycinica* 3-oxosteroid Δ^1-dehydrogenase (A0A066U7E0) stands out as the primary experimental target: it is the only enzyme with strong predicted affinity for both target substrates simultaneously, it belongs to a mechanistically well-characterised enzyme family with established structural data, and it derives from an organism not previously associated with retinal disease, reducing concern about pathogenicity.

The pipeline developed here — UniProt EC-class mining, AlphaFold structure retrieval, and high-throughput AutoDock Vina screening — is fully automated, reproducible, and extensible to additional lipofuscin substrates and wider enzyme databases (e.g., metagenome-assembled genomes from soil, sediment, and composting environments where selection pressure for degradation of oxidised lipids is highest). We anticipate that expanding the substrate set to include A2-GPE, isoA2E, and oxidised phospholipids will further enrich the candidate pool.

This work does not demonstrate enzymatic degradation of lipofuscin; it identifies the most promising bacterial enzymes to test. That step — expression, assay, and cellular delivery — remains to be done, and represents a tractable, fundable programme of experimental work with direct relevance to AMD, neurodegeneration, and the broader goal of reversing lysosomal ageing damage in post-mitotic cells.

Acknowledgements

Molecular docking was performed using AutoDock Vina executed via the Python API. Protein structures were retrieved from the AlphaFold Protein Structure Database (EMBL-EBI). Ligand structures were obtained from PubChem. Candidate enzymes were retrieved from UniProt. The literature synthesis informing target selection was generated using Claude (Anthropic). No external funding was received for this work.

Competing Interests

The author declares no competing interests.

Data and Code Availability

All docking results, ranked candidate lists, and the complete pipeline code are available at https://github.com/Klas96/lipofuscin-screen. The ranked CSV and markdown report are provided as Supplementary Data.

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