MOLECULAR ATLAS · VARIANT CARD

WFS1 Y669C — Wolframin

Tyrosine → Cysteine at position 669 in wolframin's C-terminal lumenal domain. ClinVar Likely pathogenic, associated with classical autosomal recessive Wolfram syndrome 1. AlphaMissense 0.998, DynaMut2 ΔΔG -0.41 kcal/mol (destabilising). One of the six pilot variants in the Atlas.

IDENTITY

Variant	Y669C (p.Tyrosine669Cysteine)
DNA change	c.2006A>G
Gene · Protein	WFS1 · Wolframin (890 aa)
UniProt	O76024 · WFS1_HUMAN
ClinVar accession	VCV002576526
Amino acid change	Tyrosine (Y) → Cysteine (C) — a large aromatic ring carrying a hydroxyl group replaced by a small thiol-bearing residue. Loss of aromatic packing and H-bond capacity; introduction of a reactive free thiol into the oxidizing ER lumen.

STRUCTURAL CONTEXT

AlphaFold model	AF-O76024-F1, v6
pLDDT at residue 669	**87.75** HIGH CONFIDENCE
Domain	C-terminal lumenal domain (653-869)
Position context	C-terminal lumenal domain · position 669 sits in the ER lumen in a well-folded region (pLDDT 88). The oxidizing lumenal environment is structurally relevant: any newly-introduced cysteine here can participate in aberrant disulfide chemistry.
IDR flag	No — pLDDT well above 50 threshold

Position 669 sits in wolframin's C-terminal lumenal domain (residues 653–869). The AlphaFold model places Y669 within 5 Å of GLN668 (2.5 Å), GLY670 (2.5 Å), TRP666 (3.8 Å), THR665 (4.0 Å), ALA671 (4.4 Å), and GLN667 (4.5 Å). The wild-type tyrosine ring sits in an aromatic-adjacent environment — TRP666 at 3.8 Å is consistent with edge-face π–π stacking between the Y669 phenol and the W666 indole. The hydroxyl group of the wild-type also makes plausible hydrogen-bonding contacts to GLN668 and THR665. Replacing tyrosine with cysteine at this position has three layered effects. First, the aromatic ring is gone, eliminating the π-stacking with TRP666 and the

hydrophobic surface contributed to the local fold. Second, the hydroxyl group is gone, removing two hydrogen-bond contacts. Third, a reactive free thiol is introduced into the oxidizing ER lumen — and the lumenal domain contains other cysteine residues (notably CYS673 at 3.8 Å from C690, a separate position) that could engage in aberrant disulfide formation. The new C669 is not directly in contact with another cysteine in the AlphaFold model, but disulfide chemistry in an oxidizing compartment is promiscuous. DynaMut2 returns a modest |ΔΔG| of 0.41 kcal/mol. The fold survives the substitution. The damage is mechanistic — lost aromatic packing, lost H-bonding, plus the off-pathway disulfide risk that DynaMut2's structural prediction cannot fully capture.

COMPUTATIONAL PREDICTIONS

ALPHAMISSENSE

0.998

am_class: **LPath** — threshold > 0.564

DYNAMUT2 ΔΔG

-0.41 kcal/mol

Destabilising · Job 177986259282

PLDDT (ALPHAFOLD)

87.75

high confidence

CLINICAL EVIDENCE

ClinVar classification	**LIKELY PATHOGENIC**
Review status	criteria provided, single submitter
Last evaluated	2023/08/15 00:00
Inheritance	Autosomal recessive Wolfram syndrome 1 phenotype documented in ClinVar.
WFS1 variant landscape	Y669C is 1 of ~326 pathogenic-spectrum variants in WFS1 (out of 2,243 in ClinVar)

- Wolfram syndrome 1

RESEARCH PATH DECISION TREE

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ΔΔG < 2 + binding site affected → CATEGORY 3 — docking experiments ΔΔG 2–4 → CATEGORY 2 — pharmacological chaperones ΔΔG > 4 → CATEGORY 1 — gene therapy pLDDT < 50 → CATEGORY 5 — IDR, experimental only Stable fold + functional site hit → CATEGORY 4 — site-specific docking
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Category 3/4 — Most Druggable. |ΔΔG| = 0.41 kcal/mol — fold intact. AlphaMissense 0.998 confirms severe functional consequence despite the modest structural cost.

The mechanism is layered: aromatic stacking with TRP666 broken, hydrogen bonding to GLN668/THR665 lost, and a new free thiol introduced into a compartment where it can form aberrant disulfides. Site-directed small molecules that occupy the lost aromatic packing footprint and/or block the introduced thiol are the rational therapeutic vector.

Compare with Y669H at the same position (Atlas card adjacent): Y669H preserves more of the aromatic character but introduces titratable basicity, and shows a stronger ΔΔG of -1.2 kcal/mol. The two variants together illustrate how substitution chemistry at a single position drives different mechanism and severity.

Y669C is one of the six pilot variants the Atlas was built around (R558C, A716T, G695V, L543R, A48V, Y669C). It exemplifies the lumenal domain's vulnerability to substitutions that disrupt aromatic-aromatic π-stacking — a structural feature wolframin's lumenal fold appears to depend on heavily. Drug discovery aimed at this position should target the W666 aromatic environment as the rescue surface.