1. Supplementary Tables

Supplementary Table S1. Bioinformatics tools employed for structural, evolutionary, and molecular interaction analyses of *Nicotiana tabacum* VLP, including software functions, access links, and critical parameters

Tool Name	Purpose	Link	Key Parameters/Notes
UniProt	Retrieval of N. tabacum reference proteome	https://www.uniprot.org/	Proteome ID: UP000084051
InterPro	Structural/functional annotation of uncharacterized proteins	https://www.ebi.ac.uk/interpro/	Integrates 18 databases (Pfam, SMART, etc.) and tools (TMHMM, SignalP)
Argot2.5	Functional annotation via GO term clustering	https://www.medcomp.medicina.unipd.it/Argot2-5/	Combines BLAST (homology) + HMMER (domain) hits
ExPASy-ProtParam	Physicochemical property calculation	https://web.expasy.org/protparam/	Calculates pI, MW, instability index, etc.
EMBOSS-PEPSTATS	Additional physicochemical analysis	https://www.ebi.ac.uk/Tools/seqstats/emboss_pepstats/	Computes absorption coefficients, inclusion body probability
BLASTp	Identification of VLP homologs	https://blast.ncbi.nlm.nih.gov/	90% sequence identity threshold; clustered nr database
MEGA X	Phylogenetic tree construction	https://www.megasoftware.net/	1,000 bootstrap replicates; LG+G model (BIC-selected)
I-TASSER	Ab initio 3D structure prediction	http://zhanglab.dcmb.med.umich.edu/I-TASSER/	Confidence scores (0-1); PDB threading + fragment assembly
ROBETTA	Deep learning-based structure prediction	http://robetta.bakerlab.org	Uses RoseTTAFold algorithm
AlphaFold	AI-driven structure prediction	https://alphafold.ebi.ac.uk/	AF-A0A1S4CXB2-F1 model

GalaxyRefine	3D model refinement	https://galaxy.seoklab.org/cgi-bin/submit.cgi?type=REFINE	Molecular dynamics relaxation
MOE	Molecular docking (VLP-PGN/β-glucan)	https://www.chemcomp.com/Products.htm	SiteFinder for active site prediction; OPLS_2005 force field
iMODS	Normal Mode Analysis (NMA) of docked complexes	https://imods.iqf.csic.es/	Backbone atoms (N,HA,C); 10-min execution limit
Desmond	100ns Molecular Dynamics Simulations	https://www.schrodinger.com/products/desmond/	TIP3P water model; 300K, 1 atm; RMSD/RMSF analysis
MM-GBSA (Prime)	Binding free energy calculations	Integrated in Schrödinger Suite	ΔGbind = ΔEgas + ΔGsol - TΔS; 25ns interval frames

Supplementary Table S2. Accession Numbers, Protein Names, and Plant Species Corresponding to Hypothetical and Uncharacterized Proteins Used in the Phylogenetic Analysis of VLPs from *N. tabacum* and Its Homologs. The sequences were retrieved from the NCBI Clustered Non-Redundant (nr) database via BLASTp for construction of a phylogenetic tree to study the evolutionary relationships among VLPs in Nicotiana tabacum and its homologs. The accession numbers listed provide transparency and reproducibility of the analysis.

Accession Number	Protein Name	Plant Species
A0A1S4CXB2	Vitellogenin-like protein	*Nicotiana tabacum*
XP_019236539.1	Predicted: uncharacterized protein loc109216797	*Nicotiana attenuata*
MCD7447105.1	Hypothetical protein	*Datura stramonium*
XP_059280700.1	Uncharacterized protein loc132034383	*Lycium ferocissimum*
KAJ8573882.1	Hypothetical protein k7x08_010393	*Anisodus acutangulus*
PHT60023.1	Hypothetical protein cqw23_02386	*Capsicum baccatum*
CAK9145591.1	Unnamed protein product	*Ilex paraguariensis*
KAK2990122.1	Hypothetical protein rj640_014791	*Escallonia rubra*
CDO98849.1	Unnamed protein product	*Coffea canephora*
KAK3017966.1	Hypothetical protein rj639_002723	*Escallonia herrerae*
KAI3459261.1	Hypothetical protein pfo_015924	*Paulownia fortunei*

KAI5650817.1	Hypothetical protein m9h77_36822	*Catharanthus roseus*
XP_031111088.1	Uncharacterized protein ddb_g0271670-like	*Ipomoea triloba*
KAK4737683.1	Hypothetical protein r3w88_001380	*Solanum pinnatisectum*
XP_011071438.1	Uncharacterized protein loc105156886	*Sesamum indicum*
XP_022874659.1	Uncharacterized protein loc111393383	*Olea europaea var. sylvestris*
PIM97256.1	Hypothetical protein cdl12_30275	*Handroanthus impetiginosus*
XP_057772251.1	Uncharacterized protein loc130991854	*Salvia miltiorrhiza*
TMW83782.1	Hypothetical protein ejd97_000742	*Solanum chilense*
XP_055815206.1	Uncharacterized protein loc129884976	*Solanum dulcamara*
XP_042047656.1	Mediator of rna polymerase ii transcription subunit 1-like	*Salvia splendens*
XP_059630442.1	Uncharacterized protein loc132273503	*Cornus florida*
GAA0167715.1	Hypothetical protein lier_22586	*Lithospermum erythrorhizon*
KAI3505104.1	Hypothetical protein l1887_27010	*Cichorium endivia*
RAL38543.1	Hypothetical protein dm860_002521	*Cuscuta australis*
XP_019181047.1	Predicted: uncharacterized protein loc109176080	*Ipomoea nil*
XP_009622778.1	Uncharacterized protein loc104114114 isoform x1	*Nicotiana tomentosiformis*
CAI9118842.1	Olc1v1020469c1	*Oldenlandia corymbosa var. corymbosa*
KAI3828947.1	Hypothetical protein l1987_03059	*Smallanthus sonchifolius*
CAH9080188.1	Unnamed protein product	*Cuscuta europaea*
GFQ05916.1	Hypothetical protein phja_002735600	*Phtheirospermum japonicum*
KAA8548945.1	Hypothetical protein f0562_000629	*Nyssa sinensis*
XP_051125570.1	Uncharacterized protein loc127247655	*Andrographis paniculata*
XP_052191528.1	Uncharacterized protein loc127800767	*Diospyros lotus*
KAK6161094.1	Hypothetical protein dh2020_004475	*Rehmannia glutinosa*

GER52175.1	2-oxoglutarate (2og) and fe(ii)-dependent oxygenase superfamily protein	*Striga asiatica*
XP_027116061.1	Uncharacterized protein loc113733994 isoform x1	*Coffea arabica*
KAK1418989.1	Hypothetical protein qvd17_28143	*Tagetes erecta*
KAJ0769746.1	Hypothetical protein hanlr1_chr03g0116081	*Helianthus annuus*
KAG8369926.1	Hypothetical protein bualt_bualt14g0064100	*Buddleja alternifolia*
XP_049347780.1	Uncharacterized protein loc125812311	*Solanum verrucosum*
KAD4178257.1	Hypothetical protein e3n88_26848	*Mikania micrantha*
XP_016537364.1	Uncharacterized protein ddb_g0271670	*Capsicum annuum*
KAB5537372.1	Hypothetical protein dkx38_014905	*Salix brachista*
XP_024996655.1	Uncharacterized protein ddb_g0271670-like	*Cynara cardunculus var. scolymus*
XP_023921252.1	Uncharacterized protein loc112032724 isoform x1	*Quercus suber*
XP_002270026.1	Uncharacterized protein loc100244834	*Vitis vinifera*
XP_017216725.1	Uncharacterized protein loc108194283	*Daucus carota subsp. sativus*
PSR92581.1	Osteocalcin 2 like	*Actinidia chinensis var. chinensis*
XP_002516943.2	Uncharacterized protein loc8278747	*Ricinus communis*
KAF3456241.1	Hypothetical protein fnv43_rr00891	*Rhamnella rubrinervis*
XP_047338746.1	Uncharacterized protein ddb_g0271670-like	*Impatiens glandulifera*
XP_031277209.1	Uncharacterized protein loc116135636	*Pistacia vera*
KDP27815.1	Hypothetical protein jcgz_18895	*Jatropha curcas*
XP_043628371.1	Uncharacterized protein loc122599847	*Erigeron canadensis*
KAJ4834088.1	Hypothetical protein tsubulata_016287	*Turnera subulata*
XP_044485159.1	Uncharacterized serine-rich protein c215.13-like	*Mangifera indica*
KAI3691574.1	Hypothetical protein l6452_31370	*Arctium lappa*
GEV80543.1	Hypothetical protein	*Tanacetum cinerariifolium*
XP_018810076.1	Uncharacterized protein loc108983019 isoform x2	*Juglans regia*
KAF2310950.1	Hypothetical protein gh714_018721	*Hevea brasiliensis*
KAK1390464.1	Myristoylated alanine-rich c-kinase	*Heracleum sosnowskyi*
XP_028092390.1	Uncharacterized protein loc114292589	*Camellia sinensis*

GFY85118.1	Hypothetical protein acr_03g0018920	*Actinidia rufa*
CAI9762629.1	Unnamed protein product	*Fraxinus pennsylvanica*
XP_006419538.1	Sericin 1	*Citrus x clementina*
KAH0704950.1	Hypothetical protein ky289_010026	*Solanum tuberosum*
KZV46670.1	Hypothetical protein f511_10835	*Dorcoceras hygrometricum*
KAJ4722150.1	Vitellogenin-like protein	*Melia azedarach*
KAB1213971.1	Hypothetical protein cj030_mr5g017234	*Morella rubra*
CAA0834223.1	Unknown protein	*Striga hermonthica*
XP_011003913.1	Predicted: uncharacterized protein loc105110537	*Populus euphratica*
MBA0869614.1	Hypothetical protein	*Gossypium schwendimanii*
KAI4345399.1	Hypothetical protein l6164_012527	*Bauhinia variegata*
CAK7333398.1	Unnamed protein product	*Dovyalis caffra*
GAV64143.1	Hypothetical protein cfol_v3_07661	*Cephalotus follicularis*
XP_038719976.1	Uncharacterized protein loc120012613	*Tripterygium wilfordii*
KAH7573554.1	Hypothetical protein jro89_xs03g0170600	*Xanthoceras sorbifolium*
EOY06555.1	Uncharacterized protein tcm_021236	*Theobroma cacao*

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