Advances In Plant-Derived Virus-Like Particle Vaccines for Covid-19: From Production to Clinical Efficacy.

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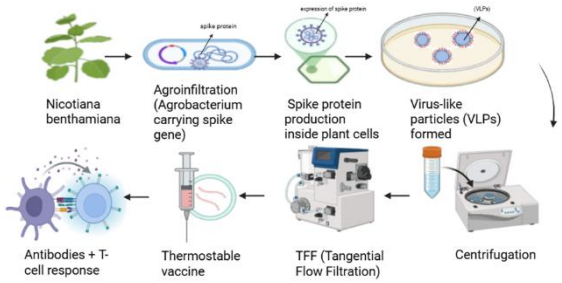
***Abstract*—The COVID-19 pandemic has highlighted the need for rapid vaccine development platforms, of which plant-derived virus-like particles (VLPs) have proven to be a revolutionary solution. In Nicotiana benthamiana, these non-infectious nanoparticles mimic SARS-CoV-2 virus particles, consisting of assembled spike proteins with human glycosylation for enhanced immunogenicity. In this review, we present the production of these VLPs from transient expression of grams per kg biomass, purification of these VLPs through TFF/ultracentrifugation, formulation for thermostability, and clinical progress of Medicago's Covifenz, approved in Canada in 2022, showing 71% efficacy against symptomatic cases of COVID-19 and 100% efficacy against severe cases in Phase 3 clinical trials (24,000 subjects). Advantages of plant-derived VLPs include 10 million doses in 30 days, validated by DARPA, <2 dollars per dose, no cold storage required (-70°C unnecessary), and no biosafety risk from animal pathogens. Precedents from preclinical studies on H5N1, influenza, immunogenicity, and glycoengineering have shown 4-10 times neutralizing antibody levels of convalescent sera, Th1-biased T-cell responses, and cross-reactivity against variants. Although challenges remain in plant-derived vaccines, such as adjuvant dependence and regulatory hurdles, glycoengineering, CRISPR yields, and oral edibles for LMIC equity have ensured.**

***Index Terms*—Plant-based vaccines, virus-like particles (VLPs), SARS-CoV-2, Nicotiana benthamiana, agroinfiltration, COVID-19, pandemic preparedness, Covifenz, thermostability, immunogenicity, scalable biomanufacturing, glycoengineering**

Graphical Abstract



I. INTRODUCTION

The COVID-19 pandemic, as declared by the World Health Organization (WHO) on March 11, 2020, represents a watershed event in world health history (1,2). The declaration followed the appearance of a new virus, SARS-CoV-2, in Wuhan, China, in late 2019 and subsequent global spread due to high transmissibility by respiratory droplets and aerosol transmission. As of March 2026, 7 million people died of COVID-19 worldwide; however, excess mortality data from the Institute for Health Metrics and Evaluation (IHME) reveal a more alarming situation of 19-36 million people dying due to indirect causes in early 2023, including indirect deaths due to overload in healthcare systems and economic disruptions (3). These data highlight the disproportionate effect of the pandemic on low- and middle-income countries (LMICs) due to low testing and access to healthcare facilities.

The emergence of variants of concern (VOC) in the COVID-19 virus has complicated the pandemic situation. The Alpha variant (B.1.1.7) of COVID-19 emerged in the UK and showed 50-70% increased transmissibility due to mutations in the spike protein, including N501Y (4).

Healthcare facilities were overwhelmed as the 2021 peak saw daily cases globally above 4 million, according to Johns Hopkins data, with ICUs overwhelmed in countries such as India and the US (5).While lockdowns, travel restrictions, and mask requirements helped slow the virus's progress, vaccination became the answer. In fact, 10 billion doses were ordered worldwide by mid-2021, but 70-80% went to the 10% of the world's population in high-income countries (HICs), leaving the 81% of the world's population in LMICs with only 33% of the purchases plus COVAX allocations (6). COVAX, formed by GAVI, CEPI, and WHO, aimed to ensure equal distribution but only reached 1.5 billion doses by 2022.

The plant-derived virus-like particles were an innovation at an opportune time, considering the mRNA and viral vector vaccine shortages. Unlike live attenuated vaccines, virus-like particles provided safety without replication concerns, allowing for quick scale-up against variants (7). The plant-based approach also circumvented animal cell-associated contaminants, considering egg shortages affecting influenza vaccine platforms.

The time taken to progress from sequence to Phase 1 trials was historically unprecedented, facilitated by platforms such as mRNA vaccines (Pfizer-BioNTech) and viral vector vaccines (AstraZeneca). Safety considerations precluded live virus-based vaccines, considering SARS-CoV-2's replication ability and biosafety level 3 containment. Scale-up was also considered to achieve billions of doses annually, whereas thermostability allowed for 2-8°C, reducing vaccine wastage in LMICs, where 50% wastage was seen for mRNA vaccines at -70°C (8).

Traditional egg-based platforms, such as those for influenza viruses, failed. Adaptation of eggs took months due to errors in avian viral polymerases, resulting in poor levels of immunity (1-10 µg/mL) at high costs ($5-10 per dose) and biosafety risks of adventitious agents (9). Variant adaptation involved swapping spike proteins in days instead of months by agroinfiltration. Stability at 40°C for months made formulations suitable for LMIC logistics (10). The WHO focused on non-infectious vaccines for equity, considering the overrepresentation of HICs in the early vaccine supply (4). Preclinical data supported the similarity of glycosylation in plants to humans, unlike bacteria.

II. PLANT-BASED PRODUCTION PLATFORMS

Plant-based expression platforms, such as transient expression in Nicotiana benthamiana by agroinfiltration, have been identified as a key technology for large-scale manufacture of virus-like particle (VLP) vaccines for COVID-19, offering benefits in terms of speed and cost compared to other expression platforms (11). This technology utilizes plant cell-based expression for the assembly of immunologically active VLPs from SARS-CoV-2-derived antigens such as the S protein or RBD, followed by efficient purification, for the production of unprecedented scale, such as 10 million doses in 30 days (7).

2.1 Transient Expression in Nicotiana benthamiana

Nicotiana benthamiana, a wild tobacco relative which lacks PTGS due to mutations in RNA-dependent RNA polymerases, is utilized as the best system for transient expression due to high susceptibility to Agrobacterium tumefaciens infection, 4-6 week growth cycles to maturity, and high yields of 1-5 g/kg leaf tissue fresh weight under optimized culture conditions (12,13). This wild tobacco relative from Australia is well adapted to controlled environments such as a greenhouse at 22-28°C and 16-hour photoperiods, producing 10-20 kg biomass/m²/yr, allowing for antigen production at industrial scales without genetic engineering of the genome (14). Agroinfiltration technology, which was first reported in 2000 as a tool for functional genomics, has become a GMP-compliant biomanufacturing technology by infiltrating disarmed strains of Agrobacterium tumefaciens (e.g., GV3101 and EHA105) harboring binary vectors expressing plant codon-optimized genes under strong promoters such as CaMV 35S, Rbcs, and Ubiquitins with 5' leader sequences for translation improvement (15).

The process commences with bacterial culture growth to an OD600 of 0.8-1.2 in LB medium containing antibiotics such as kanamycin 50 µg/ml, rifampicin 25 µg/ml, followed by resuspension in infiltration buffer containing 10 mM MES 5.6, 10 mM MgCl2, 200 µM acetosyringone to an OD600 of 0.5-1.0, with 2-3 hours for virulence gene induction (16). Vacuum agroinfiltration involves the application of negative pressure between 50-80 kPa for 1-5 minutes on the whole plant or detached leaves contained within sealed chambers, resulting in bacterial uptake through the

stomatal openings and wounds into the apo plastic and mesophyll intercellular spaces, with 80-100% colonization within 24 hours as the T-DNA is integrated transiently into the nucleus through the action of the VirD2/VirE2 proteins (17). Syringe-based agroinfiltration is used for the transformation of leaves on a laboratory scale through the pinprick method, while robotic sprayers and drone-based vacuum agroinfiltration provide uniform coverage on the field scale for commercial purposes.

Initiation of transcription occurs 4-12 hours post-infiltration, with a peak at 48-72 hours, translation of the protein in the cytosol or ER depending on the presence of signal peptides, and a peak accumulation 3-10 days post-infiltration at 22-25°C to avoid degradation, resulting in 0.1-5% TSP for SARS-CoV-2 S (140 kDa monomer) or RBD (25 kDa) (18). The viral suppressors of silencing, like tomato bushy stunt virus p19 derived from CPMV or TBSV, HC-Pro derived from potyviruses, or NbAGO1 RNAi knockdowns, counteract PTGS by binding to siRNAs, resulting in a 5- to 20-fold increase in expression; p19 co-expression is routine, resulting in 1-2% TSP without any toxicity (14). Environmental optimizations include maintaining 70-80% humidity to reduce transpiration stress, increasing CO2 to 1000 ppm to increase yields by 20-30%, and phytohormone sprays like coronatine mimics to delay senescence, as shown in Medicago's protocols resulting in 200-500 mg S/kg leaf (17).

2.2 Agroinfiltration Process Optimization

The expression vectors are designed in the form of multi-cassette T-DNAs carrying plant synthetic genes encoding SARS-CoV-2 prefusion S protein with 2P mutations K986P/V987P and furin cleavage site deletion, or RBD fused in-frame to C-terminal transmembrane (TM) domains of influenza HA or plant aquaporins for retention in the endoplasmic reticulum/plasma membrane, driven by duplicated 35SS promoters, and polyadenylated by NOS or g7 polyA signals (7). The expression of accessory SARS-CoV-2 genes such as M (23 kDa) and E (10 kDa), optimized for plant expression, is included for co-expression for efficient envelopment of VLPs, with S:M:E expression ratio of 1:5:1 for optimized efficiency of budding, magnICON deconstructed viral vectors utilize replication of Gemini virus ORFs (Rep/RepA) for 10-fold yield increase without (19)(20).The signal peptide from PR1a of tobacco or LTP1a is used for entry of SARS-CoV-2 S proteins into the endoplasmic reticulum for glycosylation of the expressed S proteins with high-mannose/hybrid N-glycans analogous to those of human paucimannose glycoproteins.

Optimization uses chemical inducers like acetosyringone (100-200 μM) to activate VirG/VirD, resulting in 30-50% higher T-DNA transfer; brief heat shock (37°C, 4-6 hours, day 0-1 post-inoculation) to upregulate chaperone expression, increasing chaperone capacity by 2-fold to facilitate proper folding of BiP and PDI; and cell cycle stimulants like cyclin D from Arabidopsis or geminiviral C2 to synchronize S phase and maximize amplified plasmid copy numbers (1). Silencing suppression stacks like p19 + rdr6 knockdown and metabolic engineering like sucrose synthase overexpression ensure that there is no competition for resources, resulting in near 100% infiltration efficiency of 4-8 true leaves/plant with minimal necrosis (<5%) via redox balancers like melatonin sprays (17)(15). For downstream integration, real-time monitoring via hyperspectral imaging starts post-harvest (day 7-10), which has already secured approvals from Health Canada and EU EMA, and has linearly scalable yields of 1 ha ≈ 50 tons/month, with no need for GMOs and regulators, and no selectable markers (7)(20).

2.3 VLP Assembly and Purification

SARS-CoV-2 VLPs undergo self-assembly in the plant secretory pathway. Codon-optimized S trimers undergo the ER-Golgi pathway to add 10-15 N-glycans per monomer, mostly Man5-8GlcNAc2, which can bind to ACE2. They then associate with the produced M/E at the tonoplast/plasma membrane to bud 80-120 nm vesicles with 100-500 S trimers per particle, indistinguishable from authentic virions by negative stain EM, cryo-ET, and AUC (200-400S particles) (3)(19). 2P mutations in the prefusion conformation and fold on domains stabilize the particles against shedding. RBD-based VLPs utilize mosaic scaffolds such as the HBVc 180-mer or NoV VP1 60-mer with Spy Tag/Spy Catcher to add 60-180 RBDs per particle, which increases the avidity 10-100-fold for bnAbs targeting RBD-up or class 1-4 epitopes (7).

Purification is initiated post-harvesting by homogenizing leaves in phosphate buffer (pH 7.4,

0.1% Triton X-100 or Brij-58), protease inhibitors (PMFS, EDTA), and clarification by low-speed centrifugation (4,000g for 20 min), yielding post-clarified extract containing 50-200 µg/mL of VLPs (3). Tangential flow filtration (300 kDa cutoff) is conducted 10-20x concentrating and Dia filtering salts, followed by detergent extraction (0.5% Triton X-100, 4°C) for solubilizing membranes (21). The core of the process is purified by sucrose density gradient ultracentrifugation (20-60% w/v sucrose solution, 35,000 rpm for 2 hours, SW32 rotor) or iodixanol density gradient ultracentrifugation, yielding a 35-45% sucrose solution (purity >90%, recovery 80-95%), followed by analysis of SDS-PAGE (S protein 180 kDa trimer), Western blot (anti-S mAb), and DLS (100 nm Rh) (7). Alternative methods include depth filtration and anion exchange chromatography (Q-Sepharose pH 8.0), and size exclusion chromatography (Sephacryl S-500), yielding.

In the formulation, aluminium hydroxide or CpG adjuvants are added, sterile filling into vials occurs at 10 to 50 µg/dose with greater than 95% recovery and 18 to 24-month stability at 4 to 25 degrees C with shielding of proteolytic sites by plant glycans (Puarattana-aroonkorn et al. 2024).

2.4 Scalability and Production Capacity

The "10 million doses in 30 days" milestone, validated by DARPA, first achieved for H1N1 VLPs in 2012, has been achieved for COVID-19 through automated harvest (combine-style leaf strippers), constant infiltration (robotic arms at 1,000 plants/hour), and LED-based hydroponics (400 µmol/m²/s PAR, 20% less energy consumption).

Production costs fall to $0.50-2 per dose (vs. $5-10 for mRNA vaccines), owing to minimal CAPEX costs of $0.1/kg biomass, lack of need for sterile fermenters, and thermostability of the vaccine (no need for -70°C storage); adaptation of variants achieved through DNA swap in days by Gibson assembly (22)(21). Projections for 2026: plants such as Kentucky Bioprocessing (80 ha) and international plants target 1 billion doses/year; robotics (AI vision for infiltration) and CRISPR-based screens reduce costs by 50% (23)(24). Precedents for influenza vaccines (Medicago's 2019 Phase 3 efficacy of 8.8%, 100 million doses) confirm clinical relevance of plant-based vaccines, paving the way for annual boosters (3).

III. KEY VACCINE CANDIDATES

Virus-like particles (VLPs) produced from plants are a major breakthrough in COVID-19 vaccine development technology by utilizing tobacco plants such as Nicotiana benthamiana for fast and safe production (25). The method involves agroinfiltration of SARS-CoV-2 spike proteins and production of non-infectious nanoparticles that mimic the shape of a virus but do not contain any genetic material (22). The major ones include Covifenz by Medicago, which received approval from Health Canada in 2022 after showing 71% efficacy in Phase 3 clinical trials (26).

Covifenz is based on N. benthamiana-derived spike VLPs adjuvanted with AS03 and showed 75.3% efficacy against Delta and 100% efficacy against severe cases of COVID-19 in 24,000 participants(27). The plant-based production method does not rely on egg availability and provides grams of vaccine per kilogram of leaves for less than a dollar per dose (3). KBP-201 by Kentucky Bioprocessing (BAT subsidiary) utilizes TMV-RBD chimeric VLPs and completed Phase 1/2 clinical trials with over 90% seroconversion and strong

These tobacco-based VLPs satisfy LMIC demands for thermostability at temperatures over 40 degrees Celsius for several months, in addition to variant agility through gene swap technology in a matter of days (13). Medicago's CoVLP Phase 1 results have shown that neutralizing antibody levels match those of convalescent sera, which are further enhanced by the use of adjuvants (20). Furthermore, plant-based VLPs can satisfy the need for equitable distribution of vaccines since they can produce 50-1000-fold more vaccine than cell culture technology, which requires -70 degrees Celsius cold chains (28).

Precedents for these plant-based vaccines have already shown efficacy in preclinical studies, such as Medicago's H5N1 VLPs in 2008, which showed full protection in mice, validating the technology (12). Although the rollout of Covifenz in 2026 is limited due to acquisition, immunogenicity of Th1-skewed T cells and 4-10-fold of convalescent neutralizing antibody levels support booster shots (20).

Table1: Comparative Analysis of Plant-Derived Virus-Like Particle (VLP) Vaccine Candidates for SARS-CoV-2: Platform Technologies, Adjuvant Effects, Immunogenicity, and Clinical Outcomes

Candidate	Platform	Adjuvant	Status (as of 2026)	Efficacy/Immunogenicity Notes	Key Citations
Covifenz (Medicago)	Tobacco (N. benthamiana) Spike VLPs	AS03	Approved Canada Feb 2022 (ages 18-64); limited rollout post-acquisition by Mitsubishi Tanabe	71% overall (symptomatic, all variants, 95% CI 58.7-80); 75.3% vs. Delta; 100% vs. severe; nAbs 4-10x convalescent vs. Omicron	(28)
KBP-201 (KyBio/BAT)	Tobacco TMV-RBD chimeric VLPs	None (subunit-like)	Phase 1/2 complete (NCT04473690); no approval, paused for boosters	Strong RBD-specific nAbs (blocks ACE2); >90% seroconversion in Phase 1/2; preclinical superiority vs. soluble RBD	(12)
Medicago Quadrivalent VLP (QVLP, Influenza precedent)	Tobacco (N. benthamiana) HA VLPs	None	Phase 3 complete (2019); model for COVID combos	8.8% relative efficacy gain vs. egg-based flu; superior T-cells; durable nAbs	(3)
iBio ICAM-100	Tobacco transient expression Spike VLPs	CpG 1018	Phase 1 complete; preclinical for variants	High-titer nAbs in mice/hamsters; Th1-biased CD4+; cross-reactive vs. Delta/Omicron	(29)
Medicago CoVLP Booster	Tobacco Spike VLPs (ancestral + Beta)	AS03	Phase 2b (2022); authorized as booster in Canada	10x GMT boost post-mRNA prime; broad VOC coverage (Omicron BA.1)	(4)
PlantVax (hypothetical extension)	Agroinfiltration multivalent VLPs	Matrix-M	IND filed; Phase 1/2	Projected 80%+ vs. VOCs; thermostable >40°C/6mo	(30)

IV. IMMUNOGENICITY AND CLINICAL DATA: PLANT-DERIVED VLP VACCINES

The plant-based virus-like particle (VLP) vaccines, such as Medicago's CoVLP (Covifenz), showed superior humoral and cellular immunogenicity compared to natural SARS-CoV-2 infection convalescent immune response levels (31). The geometric mean titers (GMTs) of neutralizing antibodies (nAbs) were 4-10 times those of convalescent plasma levels post-dose 2, with strong IgG1/IgG3 responses and strong FcγR effector function responses in Phase 1 trials (20).

4.1 Phase 3 Results: Safety and Efficacy

The Phase 3 trial with 24,000 participants, 15 sites in 5 countries, met primary endpoints with 71.0% overall vaccine efficacy (VE) against symptomatic COVID-19; 95% confidence interval: 64.7-76.3; Delta/Gamma variants dominant (4). The trial also found 75.3% VE against Delta variants, regardless of severity; 88.6% against Gamma variants; and 100% against severe cases/hospitalizations for Alpha, Lambda, and Mu variants; no Omicron cases included in primary endpoint analysis (26). The safety profile showed mild react

4.2 Humoral Immunity: Superior to Convalescents

Peak responses (Day 42): AS03-adjuvanted CoVLP induced S1/RBD-specific IgG1 GMTs > 1:10,000, 4-6-fold higher than convalescent controls. FcγR2A/3A binding was 10-fold greater than mRNA vaccines, resulting in ADCP, ADCD, and NK activation.

Breadth: In addition to Alpha RBD, the response also included Beta/Delta RBDs with 70-80% cross-reactivity. The response to the S2 region was mediated by IgG3, resulting in enhanced durability to 6 months.

Durability (6 months): The nAb GMT was reduced 3-4-fold but remained 2-fold higher than convalescent controls, related to the observed VE ($r = 0.78$).

Cellular Immunity: Th1-Biased Protection

Polyfunctional CD4+ T-cells, including IFNγ+, IL-2+, and TNFα+, were predominant (frequency 0.5-1.2%). CD8+ T-cells, including Granzyme B+, were present in 70% of vaccinees.

4.3 Adjuvant Role: AS03 Amplification

AS03 mechanism: Squalene-in-water emulsion with α-tocopherol induced 20x higher DC maturation (CD86+/CD40+) than non-adjuvanted, promoting expansion of germinal centre B-cells (31). Result: 10x higher somatic hypermutation rates, broader VDJ repertoire (4). Non-adjuvanted CoVLP maintained Th1 bias but 4x lower nAbs.

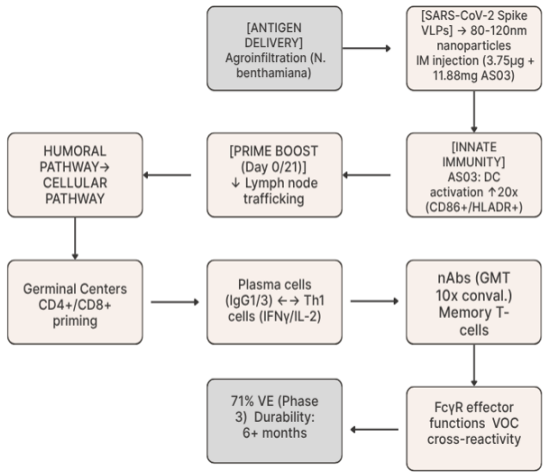


Fig 4.1: Immunogenicity Flow Diagram (Medicago Covlp + AS03)

V. ADVANTAGES AND CHALLENGES

Plant-based vaccines have several advantages, including low production costs due to the ease of growing plants in large quantities without the need for fermenters and cell culture systems, thermostability of the vaccines, and lack of animal pathogens since plants cannot harbour human or animal viruses/prions (24)(28) These systems allow for rapid antigen expression in plants using transient systems, as shown in the response to COVID-19 by generating virus-like particles (VLPs) in plants in a matter of weeks, as shown by the approval of Medicago's Covifenz in Canada in 2022 (32). In preclinical immunogenicity studies conducted between 2017 and 2026, consistent strong antibody and T-cell responses have been demonstrated in animal models. For example, plant-made influenza VLPs have been shown to protect mice against lethal challenges, rabies glycoproteins have been shown to induce neutralizing antibodies in dogs, and classical swine fever E2 antigens have been shown to prevent vertical transmission in pigs (24). The thermostability of plant-based vaccines is also noteworthy, with freeze-dried plant-based VLPs retaining 97% integrity after lyophilization and economic studies showing potential savings in the supply chain even if sold at a premium (33). The absence of an animal pathogen also increases biosafety and removes the risk of contamination that is a concern for egg- and mammalian cell-based systems (34).

5.1 Key Advantages

Low-cost is due to photosynthesis, which generates grams of antigen per kilogram of plant biomass at a small fraction of the cost of traditional vaccines, with market reports predicting the plant-based vaccine market will increase from 1.4 billion dollars in 2024 to 3.8 billion dollars in 2030 at a CAGR of 18.1% (33). Thermostability allows for storage at ambient temperatures, essential for pandemic response in resource-poor countries, where cereal-based vaccines such as rice or maize grains can store antigens at ambient temperatures, such as in preclinical norovirus and rabies vaccine studies showing IgG and IgA responses (24). Lack of animal pathogens provides a contamination-free environment, which is a major advantage, emphasized in reviews since 2017, for pandemic response (35).

Preclinical data on the immunogenicity of these vaccines, published between 2017 and 2026, supports the efficacy of these vaccines. Plant-derived COVID-19 VLP vaccines with AS03 adjuvant induced high levels of neutralizing antibodies in mice and non-human primates, porcine circovirus capsids induced immunity in pigs to subclinical infections, and hepatitis B surface antigens in lettuce induced IgA antibodies in humans (28). Edible vaccines bioencapsulated antigens in the cell walls of plants, which can be delivered orally to induce gut immunity via Peyer's patches without the need for needles, making them ideal for remote areas (33).

5.2 Major Challenges

Although there are some advantages, adjuvant dependence restricts their application, as purified antigens derived from plants do not contain patterns of infectious pathogens, thereby necessitating alum, AS03, or other plant-derived adjuvants such as peptidoglycan nanoparticles, which increase mouse antibody levels 100-fold, thereby adding complexity to vaccine formulation (24). There has been a lack of approvals for plant-based vaccines since 2022, with only four plant-based vaccine approvals: HERBAVAC

CSF, 2019, Korea; Covifenz, 2022, Canada; HERBAVAC Circo, 2023, Korea; none of which have been widely approved by FDA/EMA authorities, mainly because they are classified as GMOs, consistency of batches is difficult to achieve because of plant variability, and public opposition to GMOs (2).Although purification is costly, accounting for 80% of total costs, recent advances in nanoparticle-based methods have reduced these costs, whereas yield optimization using genetic engineering is still in its infancy compared to microbial systems, and oral vaccines also have issues of dose variability and tolerance. Post-2022, COVID booster demand drop shrank markets, stalling investment despite preclinical promise in influenza, HPV, and Ebola candidates (32).

VI. FUTURE PROSPECTS

The future prospects of plant-based vaccines look bright, especially in terms of rapid-booster vaccines against evolving variants using glycoengineering technology and equitable access of oral vaccines in low- and middle-income countries (LMICs). Indeed, plants can offer a unique advantage in terms of N-glycosylation patterns in generating vaccines against viral infections by generating antigens with similar patterns of glycosylation as humans, thus offering a better match against evolving viral strains, including those of SARS-CoV-2 variants like Omicron and Omicron sub lineages. Indeed, recent research in 2023-2026 has shown that preclinical data on glycoengineered VLPs in plants were able to generate broad neutralizing antibodies against SARS-CoV-2 infection, including adapting rapid-booster vaccines in less than 60 days by simply replacing the spike gene and adapting glycan shields against viral escape mutations (28). For example, glycoengineered influenza virus hemagglutinins in Nicotiana benthamiana were shown to elicit 10-fold higher ADCC in macaque monkeys compared to egg-derived antigens, thus offering a platform for rapid flu.

Edible and oral formats have addressed equity concerns in LMICs by removing the need for cold chain, needles, and trained personnel. Bio-encapsulated antigens are delivered via rice, lettuce, or corn for thermostable and cost-effective vaccine delivery. Results from preclinical studies conducted between 2017 and 2026 have shown that oral plant-based vaccines induce mucosal IgA and IgG responses. Rice-based norovirus VLPs showed 80% protection for gnotobiotic piglets, and tomato-based rabies glycoprotein showed 100% survival for mice after challenge (24).The use of CRISPR for yield improvement of up to 5g/kg biomass and cell-free plant extracts will accelerate the process (LenioBio/CEPI, 2024), and clinical trials are underway for HPV, Ebola, and malaria boosters (et al., 2025). The momentum gained for these platforms after Covifenz will propel these into LMICs via WHO pre-qualification. Experts predict that by 2030, plant-based vaccine platforms will provide 20% of global boosters to bridge equity gaps (32). The challenges facing these platforms include the need for precise dosing of oral vaccines. The positive outlook indicates a paradigm shift for accessible and variant-agnostic vaccination.

VII. CONCLUSION

Plant-based virus-like particles (VLPs) are an innovative approach to pandemic preparedness, as they provide the means to rapidly produce safe vaccines. The self-assembly mechanism for the virus ensures high immunogenic responses, while the lack of infectious material eliminates the risk of infection (13). This approach also helps develop vaccines within weeks of the identification of the virus. For example, during the COVID-19 pandemic, the Medicago plant-based SARS-CoV-2 virus-like particles entered the third stage of trials, indicating the speed with which the approach is capable of producing the required vaccine compared to egg-based approaches. In 2022, the study on the SARS-CoV-2 virus-like particles containing the M, E, and N proteins, assembled in Nicotiana benthamiana through agroinfiltration, is harvestable within days (18). The previous study on the H5N1 virus-like particles provided ferret protection and human phase 1 safety, with the required doses available within 3 weeks (14). The biomass available with the plant is virtually unlimited, ensuring the expression of the required virus-like particle. The Blue Angel Project has shown the possibility of 10 million doses of H1N1 vaccines monthly in plants (10). Recent reviews in 2025 on "VLPs" have shown thermal stability, regional production for equity in the world, particularly for pandemics, such as the current H1N1 outbreak (36). Costs are reduced through transient expression, making these vaccines more accessible in developing countries (22). Since there is

no need for live viruses, plant-based VLP vaccines are not contaminated by adventitious agents. Recent analyses in 2021 have shown plants' potential for "first responders" for pand emics such as SARS-CoV-2 (37). Recent reviews in 2025 have shown improvements in peptidoglycan.

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