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PRESS RELEASE

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Scientists Demonstrate Pre-clinical Proof of Concept for Next-Gen DNA Delivery Technology

Findings show potential for new lipid-based delivery formulations as a platform technology for immunization

PHILADELPHIA — (March 21, 2025) — Scientists in The Wistar Institute lab of **David B. Weiner, Ph.D.**, in collaboration with scientists in the laboratory of Norbert Pardi, Ph.D., at the University of Pennsylvania Perelman School of Medicine and at the Pennsylvania-headquartered biotechnology company INOVIO, described a next-generation vaccination technology that combines plasmid DNA with a lipid nanoparticle (LNP) delivery system. Their findings are published in *Cell Reports Medicine* in the paper, "Modulation of lipid nanoparticle-formulated plasmid DNA drives innate immune activation promoting adaptive immunity."

David Weiner, Wistar executive vice president and W.W. Smith Charitable Trust Distinguished Professor in Cancer Research, is a leading expert in the field of DNA vaccines. In the study led by Weiner lab doctoral student **Nicholas Tursi**, researchers aimed to study how to improve lipid-based formulations to better incorporate and deliver DNA payloads for immunization.

Lipid-based approaches, including LNPs, have successfully formulated and delivered various forms of RNA as well as formulating proteins as drugs in several marketed products. However, developing such formulations using DNA has previously not shown the same stability or efficacy. Tursi et al. studied how to modify lipid-based formulations that would effectively stabilize DNA in LNPs, which would simplify their delivery and improve vaccine-induced immunity. DNA has unique properties relative to RNA, including its large size and double stranded nature, which has previously been a hurdle for creating stable and consistent lipid-based DNA formulations.

DNA vaccines have been traditionally delivered using devices, which enable highly efficient uptake of DNA into cells at the injection site and potent T cell immunity against important disease targets. Utilizing an LNP formulation for DNA vaccines could potentially enable administration by needle and

syringe and potentially enhance humoral immunity, which could provide an additional tool within the DNA vaccine toolkit.

Using a model DNA-LNP expressing influenza hemagglutinin (HA), the team examined how to modulate the formulation of DNA within LNPs to improve particle assembly and stability for direct injection. HA DNA-LNPs formulated at higher N/P ratios—the relationship between the lipid nanoparticle and the larger DNA backbone—led to an improved particle profile, smaller particle size, with an improved generation of immune responses.

The study highlights some of the mechanisms of immunity that are conferred by DNA-LNPs. The team showed that these DNA-LNPs demonstrate a unique way of priming the immune system compared to mRNA and protein-in-adjuvant formulations. The DNA-LNP induced a unique activation pattern of innate immune populations—cells that respond early in the development of a protective immune response. The team next examined whether HA DNA-LNPs could induce strong and consistent adaptive immunity—the arm of the immune system responsible for long lived T cell and antibody responses. Relative to benchmark mRNA and protein-in-adjuvant vaccines, HA DNA-LNPs induced robust antibody and T cell responses after a single dose. Importantly, these responses were durable, with memory responses in small animals persisting beyond a year after immunization. The team also examined the immunogenicity of HA DNA-LNPs in a rabbit model, where they observed strong T cell and antibody responses that persisted into the memory phase.

Finally, the team examined whether DNA-LNP vaccines could be protective in a live SARS-CoV-2 challenge model. The team utilized a DNA-LNP vaccine expressing the SARS-CoV-2 spike protein and demonstrated that a single immunization with the spike DNA-LNP successfully prevented morbidity and mortality from challenge.

This study supports the continued development DNA-LNP vaccines as a unique vaccination modality. The ability for this approach to trigger strong, long-lasting immune responses highlights its potential to complement existing approaches or be potentially developed as next-generation immunization platform.

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