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Influenza Virus Characteristics in Department of Defense Populations, 2024–2025

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Abstract

The influenza virus constantly evolves through antigenic shift and drift, requiring annual review to inform the development of seasonal influenza vaccines. The Department of Defense Global Respiratory Pathogen Surveillance Program and Global Emerging Infections Surveillance program–funded partner laboratories perform routine respiratory pathogen surveillance across a wide-reaching global network of service members and their beneficiaries, U.S. civilians, and some foreign national populations. This report describes the influenza viruses circulating during the 2024–2025 influenza season.

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Introduction

The influenza virus is a recurring threat to global public health, causing 290,000–650,000 deaths annually.[1] Influenza virus constantly evolves through antigenic shift and drift, requiring annual review to inform development of seasonal influenza vaccines. Global surveillance systems detect the most prevalent virus strains circulating. Characterization of these circulating strains is critical to preventing and responding to outbreaks, evaluating infection mechanisms, understanding viral evolution, and selecting of strains for vaccine development. Seasonal vaccination is widely considered the main strategy to protect against influenza infection and reduce disease severity. The Department of Defense (DoD) Global Respiratory Pathogen Surveillance Program and the Global Emerging Infections Surveillance program–funded partner laboratories perform routine respiratory pathogen surveillance across a wide-reaching global network of service members and their beneficiaries, U.S. civilians, and some foreign national populations. The DoD service member population is highly vaccinated[2] and widely distributed across the globe. DoD laboratory surveillance helps to identify samples for sequencing and advanced characterization that may differ from other global surveillance. Since 1979, U.S. findings, including those from DoD (when available), have been shared and discussed during an open session convened by the U.S. Food and Drug Administration's Vaccines and Related Biological Products Advisory Committee to make recommendations on the selection of strains to be included in influenza vaccines. The objective of this report is to describe the influenza viruses circulating during the 2024–2025 influenza season.

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Surveillance and Outcomes

Through DoD's network of more than 115 sentinel and laboratory partners, specimens were collected and tested for influenza on real-time polymerase chain reaction platforms, multiplex pathogen panels, and/or viral cultures. Once detected, these viruses were characterized through type-specific whole-genome sequencing and high-throughput imaging neutralization tests. The high-throughput imaging neutralization tests tested the ability of ferret antisera generated against vaccine and candidate vaccine viruses to neutralize the diversity of influenza viruses from our sentinel network. A reduction in neutralization titers generally indicates a decreased match between vaccine-derived antibodies and circulating viruses.

We characterized 1341 influenza viruses between August 2024 and February 2025. The distribution of these viruses was 55% influenza A virus subtype H3N2 (A(H3N2)), 40% pandemic influenza A virus subtype H1N1 (A(H1N1)pdm09), and 6.2% influenza B/Victoria lineage viruses, with the remaining 2.7% as undefined influenza A or B type viruses. Within the DoD network, more A(H3N2) was observed in North America, while A(H1N1)pdm09 predominated in other regions, particularly Asia. These results were consistent with data presented at the World Health Organization northern hemisphere (NH) vaccine selection meeting.[3] For A(H1N1)pdm09, the 5a.2a clade, which was first characterized in August 2021,[4] represented 63.7% of the A(H1N1)pdm09 cases (Table 1), with the remainder in the 5a.2a.1 clade, with its recently defined subclades C.1.9 (HA1: K169Q) and C.1.9.3 (HA1: S83P). Within clade 5a.2a.1, in which the egg (A/Victoria/4897/2022) and cell-based (A/Wisconsin/67/2022) components of the 2024–2025 vaccine strains reside, an expansion of D subclade viruses (HA1: T216A) was observed, including a notable increase in the D.3 (HA1: T120A) and D.5 (HA1: R45K) subclades during peak influenza positivity in January and February 2025. Ferret antisera against the 2024–2025 A(H1N1)pdm09 vaccine components exhibited protective neutralization titers against both 5a.2a and 5a.2a.1 circulating viruses.

For A(H3N2), we observed the 2a.3a.1 clade continued to expand, with several amino acid substitutions further diversifying the J.2 (HA1: K276E) subclade (Fig. 1, Table 1). Only 10 (1.5%) viruses outside of J.2 were observed. These diversifying substitutions include a series of glycosylation site losses that likely contribute to the ongoing antigenic drift of the A(H3N2) diversity, including at HA1:N8K, HA1:T135K, and HA1:N158K. The majority of our N158K substitutions occurred within a subclade defined by S145N found in the newly recommended 2025–2026 NH vaccine strain selection (egg component A/Croatia/10136RV/2023; cell component A/District of Columbia/27/2023).[3]

We observed high titers with A/District of Columbia/27/2023–derived antisera against current DoD

Table 1. Distribution of Influenza Subclade Diversity, by Influenza A Subtype and B/Victoria Lineage Virus, in the Department of Defense Network, 2024–2025

Subtype/Lineage	Clade	Subclade	No.	% of Total Subtype*	Defining Substitutions†
A(H1N1)pdm09	5a.2a	C.1.9	124	29.4	K169Q
		C.1.9.1	23	5.5	P137S, K169Q
		C.1.9.3	108	25.6	S83P, K169Q
	5a.2a.1‡	D.3	91	21.6	T120A
		D.5	40	9.5	R45K
A(H3N2)	2a.3a.1§	J.2¶	591	88.5	K276E‖
		J.2.2	53	7.9	S124N
B/Victoria	V1A.3a.2	C.5	8	9.3	
		C.5.1	37	43.0	
		C.5.6	22	25.6	
		C.5.7	18	20.9	

* Only showing subclades representing >5% of the total diversity for a given subtype (therefore, percentages do not sum to 100%).
† All substitutions are referencing changes in the hemagglutinin subunit 1.
‡ Vaccine strains A/Wisconsin/67/2022 (cell) and A/Victoria/4897/2022 (egg) selected February 2023.
§ Clade of previous 2024–2025 northern hemisphere selection A/Thailand/08/2022 (egg) and A/Massachusetts/18/2022 (cell).
¶ Subclade of selected 2025–2026 northern hemisphere strain selection A/Croatia/10136RV/2023 (egg) and A/District of Columbia/27/2023 (cell).
‖ Additional recent diversifying substitutions within J.2 include N8D(-CHO), T135K (-CHO), S145N, and N158K.

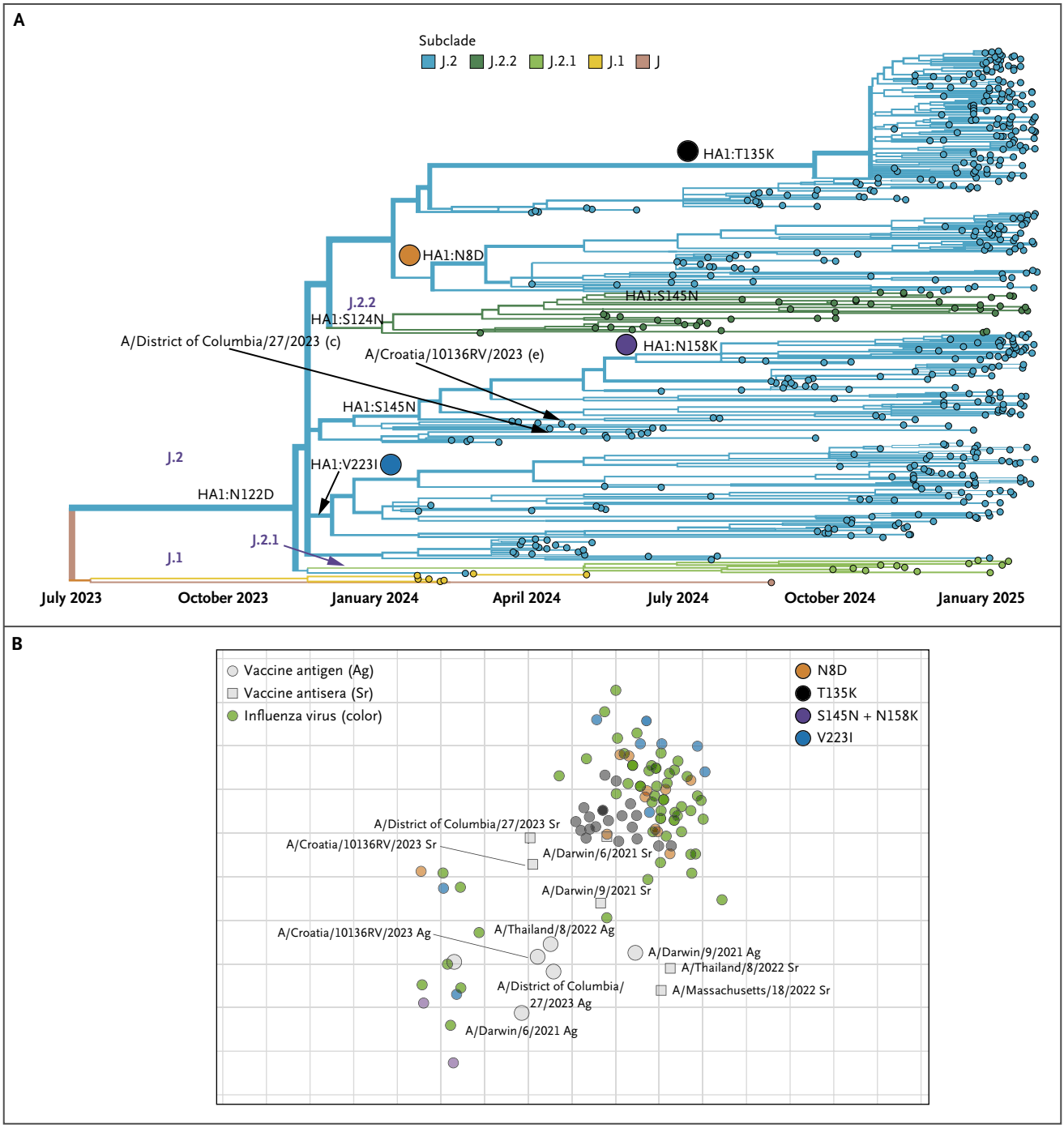

Figure 1. Phylogenetic Tree of Hemagglutinin Gene Segment Sequences for Influenza A(H3N2) and Corresponding Antigenic Cartography Map.

Panel A displays the phylogenetic tree of hemagglutinin gene segment sequences for influenza A(H3N2) in the Department of Defense network during 2024–2025. Panel B displays the corresponding antigenic cartography map showing neutralization of circulating A(H3N2) viruses by ferret antisera raised against the corresponding vaccine. A four-fold change is the standard and threshold used to determine if a new virus is considerably different (i.e., whether immunity is sufficient to provide protection). The vertical and horizontal axes (Panel B) represent antigenic distance, where the spacing between grid lines is one unit of antigenic distance. One unit of antigenic distance corresponds to a twofold dilution of antisera in the high-throughput imaging neutralization test assay. Gray circles and squares represent vaccine antigens (circle) and vaccine antisera (square), and circles of other colors represent 2024–2025 influenza viruses. Substitutions of interest identified in the viruses (Panels A and B) are in orange, black, purple, and blue circles. Viruses where substitutions of interest were not identified (Panel B) are displayed in green. Ag denotes vaccine antigen; HA, hemagglutinin; and Sr, vaccine antisera.

A(H3N2) diversity, but 35.2% (38/108 isolates) showed eightfold or greater reductions in titers when compared with homologous A/District of Columbia/27/2023 titers. In addition, 16.2% (18/111) of DoD viruses showed eightfold reduction titers against 2024–2025 cell component A/Massachusetts/18/2022 antisera when compared with the homologous titer. However, no viruses displayed eightfold reductions in neutralizing titers (0/118) against the egg-based A/Croatia/10136RV/2023 antisera when compared with the homologous A/Croatia/10136RV/2023 virus. A/Croatia/10136RV/2023 performed better than A/Thailand/08/2022 (40/111) at neutralizing recent diversity.

For influenza B viruses, we noted the continued absence of influenza B/Yamagata lineage viruses since March 2020. All genetic diversity was consistent with global trends showing a set of subclades of the C.5 (HA1:D197E) clade predominating globally. Antisera against the B/Austria/1359417/2021 egg and cell components of the 2024–2025 vaccine and recommended 2025–2026 vaccine continued to show neutralization of all B/Victoria DoD isolates within a fourfold change of the B/Austria/1359417/2021 homologous titers.

Conclusions

The DoD network performs influenza global surveillance initiatives in concert with national and international global health partners annually. These virologic data contextualize other DoD initiatives, which include vaccine effectiveness (VE) studies among active component service members (ACSM) and military health care system beneficiaries. Recent DoD reports[5,6] show the 2024–2025 midseason VE estimates were low against laboratory-confirmed outpatient influenza A (ACSM: 14%, 95% confidence interval [CI], 5 to 22%); beneficiaries: 25%, 95% CI, 1 to 44%). However, subtype-specific VE estimates in both groups showed moderate midseason protection against A(H1N1)pdm09 (ACSM: 41%, 95% CI, 14 to 60%; beneficiaries: 58%, 95% CI, 31 to 74%) and A(H3N2) (ACSM: 50%, 95% CI, 36 to 60%; beneficiaries: 42%, 95% CI, 14 to 62%).

The DoD data on circulating influenza virus characteristics and VE support the World Health Organization NH 2025–2026 influenza vaccine strain selections, which were to continue the 2024–2025 component of A(H1N1)pdm09 and B/Victoria while updating the A(H3N2) strain components.

Disclosures

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