



T-Cell Mediated Immunity Following Sequential Heterologous Vaccination Regimens

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Abstract

Sequential heterologous vaccination, in which a prime dose from one platform is followed by a boost from a different platform, moved from an emergency improvisation during the COVID-19 pandemic into standing public health guidance in several countries, and generated an unusually large, well-controlled body of human data on how platform-switching shapes T-cell mediated immunity. This short communication synthesizes findings from the Com-COV platform trials, independent European heterologous-schedule cohorts, the CombiVacS and COV-BOOST booster trials, and the T-cell epitope and variant-cross-reactivity literature underpinning them. Across these studies, heterologous regimens combining an adenoviral-vector prime with an mRNA boost, or an mRNA boost after single-dose Ad26.COV2.S priming, consistently induced spike-specific CD4+ and CD8+ T-cell responses that were comparable to or larger than those from homologous schedules, generally without a consistent tolerability penalty once the full regimen was considered. Mechanistic and variant-cross-reactivity data suggest this advantage reflects a broader, multi-epitope T-cell repertoire that is less vulnerable to spike mutations than the neutralizing-antibody response. We conclude that heterologous prime-boost vaccination is a well-evidenced strategy for eliciting robust cellular immunity, while noting that most available evidence covers only months-long follow-up and a limited set of platform combinations.

Keywords: heterologous vaccination; T-cell immunity; prime-boost; SARS-CoV-2 vaccines; cellular immunogenicity

1. Introduction

Heterologous vaccination -- priming with one vaccine platform and boosting with a different one -- moved from a niche immunological curiosity to a matter of public health policy almost overnight during the COVID-19 pandemic, when adenoviral-vector supply constraints and, later, rare thrombotic events associated with ChAdOx1 nCoV-19 forced several national programmes to offer an mRNA vaccine as the second dose to people who had already received a viral-vector prime [1]. What began as a pragmatic workaround turned out to reopen a much older question in vaccine immunology: does switching platforms between doses change not just how much antibody a person makes, but what kind of T-cell response they end up with. Mouse and non-human-primate work on heterologous prime-boost regimens, going back well before SARS-CoV-2, had already shown that combining a DNA or vectored prime with a different vectored or protein boost tends to favour broader and more durable T-cell memory than repeating the same vector twice, largely because anti-vector immunity generated by the first dose blunts the effectiveness of a second dose from the same vector [2]. The pandemic gave this mechanism an unusually large, well-instrumented human testbed.

This short communication synthesizes what a cluster of randomised trials and cohort studies published between 2021 and 2022 collectively show about T-cell mediated immunity after sequential heterologous vaccination, with COVID-19 vaccines as the dominant, best-characterised example. We are not reporting new experimental data; we are drawing together results from the Com-COV platform trials, the German and Swedish heterologous-schedule cohorts, the CombiVacS and COV-BOOST booster trials, and the T-cell epitope-mapping literature that these clinical results sit on top of, to ask a narrow, practically important question: when a heterologous regimen is used, does the resulting CD4+ and CD8+ T-cell response differ meaningfully from what a homologous regimen produces, and does that difference hold up against the antigenic drift of new variants.

2. Immunological Rationale for Heterologous Prime-Boost Regimens

The theoretical case for heterologous prime-boost regimens rests on a fairly simple observation: a viral-vector vaccine generates immunity not only against

the encoded antigen but also against the vector itself, and that anti-vector immunity -- neutralizing antibodies and vector-specific T cells -- can dampen the take of a second dose delivered by the same vector [2]. Switching to a different vector, or to a fundamentally different modality such as an mRNA-lipid-nanoparticle platform, sidesteps this ceiling and lets the second dose re-expose the immune system to the target antigen through a genuinely novel delivery context, which tends to recruit and expand a broader repertoire of antigen-specific T-cell clones rather than simply re-stimulating the same population that responded to the prime.

For SARS-CoV-2 specifically, the antigen of interest across nearly every widely deployed vaccine is the spike glycoprotein, which meant that heterologous schedules combining ChAdOx1 nCoV-19 (a chimpanzee adenoviral vector), BNT162b2 or mRNA-1273 (lipid-nanoparticle-encapsulated mRNA), and Ad26.COV2.S (a human adenoviral vector) were, immunologically, tests of platform-switching against a constant antigen rather than tests of different antigens altogether -- a relatively clean natural experiment. Pre-pandemic work characterizing the human T-cell response to coronavirus antigens, including cross-reactive memory T cells found in some SARS-CoV-2-unexposed individuals, established both the epitope landscape that vaccine-induced T cells would need to cover and the methodology (IFN-gamma ELISpot and intracellular cytokine staining against spike peptide pools) that the heterologous-schedule trials would go on to use almost uniformly [3]. That methodological consistency across studies is part of why the heterologous-schedule literature is unusually easy to compare across independent research groups and countries.

3. Antibody and Cellular Immune Responses to Heterologous ChAdOx1/mRNA Schedules

The largest and most influential platform trial in this space, the UK's Com-COV study, randomised participants to homologous and heterologous combinations of ChAdOx1 nCoV-19 and BNT162b2 given four weeks apart and reported that heterologous schedules, in either order, were more reactogenic than homologous schedules but generated antibody and cellular immune responses that met the trial's non-inferiority criteria relative to the standard homologous ChAdOx1 course, with the

ChAdOx1-prime/BNT162b2-boost combination in particular inducing higher spike-specific antibody and T-cell responses than homologous ChAdOx1/ChAdOx1 [4]. A planned follow-up analysis from the same trial examined how the interval between doses shaped these outcomes and found that, as with homologous schedules, extending the prime-boost interval improved peak immunological response for several of the heterologous combinations, indicating that the platform-switching effect and the interval effect are at least partly independent levers that vaccination programmes can tune separately [5].

Table 1: Representative trials and cohort studies of heterologous COVID-19 vaccination regimens, their comparator arm, and the T-cell/antibody outcome each cited study reports. [4,7,6,8,9,16,15].

Regimen	Source	Comparator	Reported Outcome
ChAdOx1 prime / BNT162b2 boost	Liu et al. (Com-COV) [4]	Homologous ChAdOx1/ChAdOx1	Higher spike-specific antibody and IFN-gamma ELISpot T-cell responses; non-inferior overall
ChAdOx1 prime / BNT162b2 boost	Barros-Martins et al. [7]	Homologous ChAdOx1/ChAdOx1	Higher spike-specific CD4+/CD8+ T-cell frequencies and neutralizing titres, including against Beta
ChAdOx1 prime / BNT162b2 boost	Hillus et al. [6]	Homologous ChAdOx1 and homologous BNT162b2	Higher anti-spike IgG; systemic reactions after boost in 48% vs 65% (homologous BNT162b2) and 86% (ChAdOx1 prime)
ChAdOx1 prime / mRNA-1273 boost	Normark et al. [8]	Homologous ChAdOx1/ChAdOx1	Higher binding and neutralizing antibody titres
ChAdOx1 / BNT162b2 (CombiVacS)	Borobia et al. [9]	ChAdOx1-primed, no second dose yet	Marked rise in anti-RBD IgG and IFN-gamma-producing spike-specific T cells after boost
Ad26.COV2.S / mRNA boost (SWITCH)	Sablierolle et al. [16]	Homologous Ad26.COV2.S booster	Higher spike-specific T-cell and antibody responses than homologous boosting
Primary series + 7 boosters (COV-BOOST)	Munro et al. [15]	Non-boosted control	All boosters increased spike-specific T-cell and antibody responses; heterologous boosters generally comparable or superior to homologous

Two independent European cohort studies reached compatible conclusions using different populations and slightly different assay platforms. A German cohort study of healthcare workers who received ChAdOx1 followed by BNT162b2 found that this heterologous schedule produced markedly higher anti-spike IgG concentrations and neutralizing antibody titres than a homologous ChAdOx1/ChAdOx1 schedule, and was,

notably, no more reactogenic overall than a homologous BNT162b2 schedule after the second dose: in that cohort's interim reactogenicity analysis, only 48% of heterologous-boost recipients reported systemic symptoms after their second dose, compared with 65% of participants receiving a second BNT162b2 dose in a homologous schedule and 86% after the initial ChAdOx1 prime dose itself [6]. A separate German group working with flow-cytometry-based T-cell phenotyping on a comparable ChAdOx1/BNT162b2 cohort reported that the heterologous schedule induced higher frequencies of spike-specific CD4+ and CD8+ T cells, alongside higher neutralizing titres against the ancestral strain and against the Beta variant, than homologous ChAdOx1 vaccination [7]. A Swedish cohort study using a ChAdOx1-prime/mRNA-1273-boost combination reported the same directional result with a different mRNA vaccine as the boost, reinforcing that the effect tracks with switching away from the adenoviral platform rather than with any single mRNA product [8]. Spain's CombiVacS trial, which specifically enrolled ChAdOx1-primed adults and randomised them to a BNT162b2 boost or no immediate second dose, showed a substantial rise in anti-receptor-binding-domain IgG and in the frequency of interferon-gamma-producing spike-specific T cells after the heterologous boost, data that were influential in several European countries' decisions to formally recommend heterologous schedules for adults who had already received one ChAdOx1 dose [9].

Mechanistic work on mRNA vaccination on its own helps explain why the boosting dose matters so much for the T-cell compartment specifically. Longitudinal immune monitoring after BNT162b2 vaccination found that a rapid, coordinated induction of antigen-specific CD4+ T cells after the first dose was associated with the subsequent quality of both the antibody response and the CD8+ T-cell response, suggesting that the priming event sets a trajectory that later doses, and later platform choices, build on rather than override [10]. Direct immunogenicity work on BNT162b2 itself confirmed that the vaccine induces a genuinely poly-specific T-cell response spanning multiple spike epitopes rather than a narrow response to one dominant epitope, a breadth property that later work would show matters considerably once variants begin to accumulate spike mutations [11].

4. T-Cell Breadth and Cross-Reactivity Against Variants

The practical reason T-cell responses to heterologous regimens attracted sustained attention beyond the initial immunogenicity trials is that T-cell epitopes, unlike the handful of antibody-neutralizing epitopes concentrated on the receptor-binding domain, are distributed across the entire spike protein and, for CD8+ T cells in particular, are restricted by a wide range of individual HLA alleles. This structural feature makes the T-cell compartment intrinsically harder for a small number of point mutations to escape wholesale. A study directly comparing convalescent and vaccinated donors' immune responses against SARS-CoV-2 variants of concern found that neutralizing antibody activity against Alpha, Beta, and Gamma variants was measurably reduced relative to the ancestral strain, while CD4+ and CD8+ T-cell responses against the same variants were largely preserved [12]. A later, more comprehensive analysis extended this comparison through Omicron and found that vaccine-induced T-cell memory retained the substantial majority of its reactivity against Omicron's spike sequence even as neutralizing antibody titres against that variant dropped sharply, a pattern the authors attributed to the breadth of epitopes targeted by vaccine-primed T cells rather than to any single dominant, escape-prone epitope [13].

This distinction between antibody and T-cell durability against variants is where the heterologous-regimen literature connects most directly back to disease outcomes rather than just laboratory correlates. Work in a rhesus macaque SARS-CoV-2 challenge model found that CD8+ T-cell depletion partially abrogated the protection conferred by prior vaccination or infection, particularly when antibody titres were sub-optimal, providing direct causal evidence -- something the observational human immunogenicity studies cannot by themselves provide -- that vaccine-induced T cells contribute meaningfully to protection and are not merely a correlate that travels alongside the antibody response [14]. Read together with the breadth data, this argues that a heterologous regimen's tendency to produce a broader, multi-epitope T-cell response is not an immunological curiosity but a property with a plausible protective mechanism attached to it.

5. Reactogenicity, Booster Strategies, and Practical Adoption

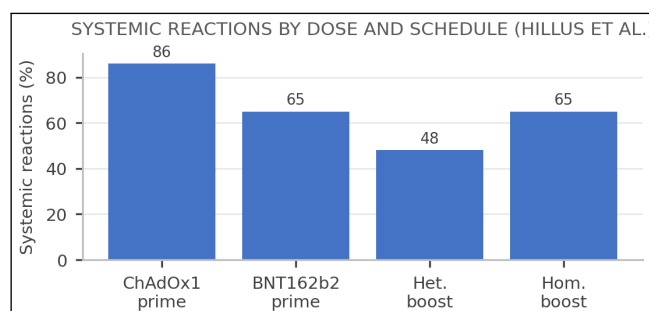


Figure 1: Proportion of participants reporting systemic reactions after each vaccination in a German cohort study of the ChAdOx1 nCoV-19/BNT162b2 heterologous prime-boost schedule. Het. boost = heterologous ChAdOx1/BNT162b2 second dose; Hom. boost = homologous BNT162b2/BNT162b2 second dose. The heterologous booster was better tolerated than either the ChAdOx1 prime dose or the homologous BNT162b2 booster [6].

Once heterologous priming schedules had been established as immunogenic and reasonably well tolerated, the natural extension was to ask whether platform-switching also improved third-dose booster responses among people who had already completed a full primary course, and here the evidence is if anything more consistent. The UK's COV-BOOST trial systematically evaluated seven different vaccines as a third-dose booster in adults who had completed a two-dose primary course of either ChAdOx1 or BNT162b2, and found that every candidate booster increased spike-specific antibody and T-cell responses relative to a control, with boosters using a different platform than the primary course generally producing responses at least as strong as, and in several combinations stronger than, a homologous third dose [15]. A separate Dutch trial, SWITCH, focused specifically on adults primed with the single-dose Ad26.COV2.S vaccine and found that boosting with either mRNA vaccine produced substantially higher spike-specific T-cell and antibody responses than a homologous Ad26.COV2.S booster, a result that was influential in several countries moving away from same-platform boosting for Ad26.COV2.S recipients specifically [16]. A large US platform trial extended the mix-and-match logic across essentially every combination of the three major US-authorized vaccines as both primary series and booster, reinforcing that heterologous boosting is broadly, rather than narrowly, advantageous across platform combinations [17].

The reactogenicity data are worth foregrounding because they run counter to an intuitive worry that switching platforms would necessarily mean a rougher second dose. In the German cohort discussed above, the heterologous ChAdOx1/BNT162b2 schedule's second

dose was in fact better tolerated, by self-reported systemic symptoms, than either a repeat ChAdOx1 dose or a homologous BNT162b2 second dose [6]. That combination of comparable-or-better tolerability and comparable-or-better cellular immunogenicity is a large part of why heterologous schedules moved so quickly from an emergency improvisation into standing public health guidance in multiple countries.

6. Conclusion

Taken across the Com-COV platform trials, the independent European and North American cohort and booster studies, and the T-cell epitope and variant-cross-reactivity literature they sit on top of, the evidence supports a fairly specific and consistent claim: sequential heterologous vaccination, and specifically the practice of priming with a viral-vector vaccine and boosting with an mRNA vaccine or vice versa, produces spike-specific CD4+ and CD8+ T-cell responses that are at least comparable to, and in several well-controlled comparisons measurably broader or larger than, those produced by repeating the same platform twice, without a consistent tolerability penalty once the whole regimen rather than the boosting dose alone is considered. The mechanistic story -- platform-switching avoiding anti-vector immunity and recruiting a broader epitope repertoire -- is well supported by both older prime-boost immunology and the newer variant-cross-reactivity data showing that this broader T-cell response holds up better against spike mutations than the antibody response does. What remains less settled is how far this generalizes beyond the specific platform combinations tested at scale during the pandemic, and how durable the T-cell advantage of heterologous priming proves to be over the multi-year timescales relevant to routine immunization schedules rather than the months-long follow-up windows most of these studies report; that longer-horizon evidence base is still being built.

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Conflict of Interest

The authors declare that they have no competing interests relevant to this work.

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Ethical Approval

Not applicable. As a literature synthesis, this article did not involve new human or animal subjects or primary data collection.

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