VIROLOGY

HIV not as simple as one, two, three

The HIV-1 envelope protein is the target of antibodies that neutralize the virus. A fresh look into the conformational states of this protein relaunches the quest to identify those relevant for vaccine design. SEE LETTER P.415

ALEXANDRA TRKOLA

The first step of infection of host cells by HIV-1 is the binding of the envelope (Env) glycoprotein, which is expressed on the surface of the virus, to cellular receptors. Env is a metastable assembly of three heterodimeric complexes that form a trimer. Lu *et al.*[1] present thought-provoking data on page 415 that relaunch the search for the native conformational states of the Env trimer.

Before binding to cellular receptors, the Env trimer is in a closed (also referred to as native) conformational state. This conformation masks certain domains that are essential for the virus to enter cells, but that would otherwise be targeted by Env-specific neutralizing antibodies generated by the host's immune system in response to HIV-1 infection. Interaction with cellular receptors — first CD4, then a co-receptor — triggers conformational changes that open up the Env trimer and pave the way for the fusion of viral- and host-cell membranes[2] (Fig. 1). Defining the diverse structures of the Env trimer is key to understanding the host's neutralizing-antibody response and to developing vaccines based on Env-derived immunogens — the proteins that stimulate the production of protective antibodies.

Despite being shielded from attacks by most neutralizing antibodies, the native trimer is potently targeted by broadly neutralizing antibodies (bnAbs). These antibodies have exceptional neutralization breadth; that is, they are active against a wide spectrum of HIV-1 subtypes and variants that emerge when the gene that encodes Env mutates. These antibodies have been investigated in HIV-vaccine development, but so far no vaccine has been able to promote a bnAb response in animals or humans[2–4]. The search is on for Env-derived immunogens that can successfully trigger bnAb activity. Resolving the structure of the native Env trimer is key, because this is the entity that is present before the virus binds to and enters cells. This might also be the immunogen that triggers bnAb production in people infected with HIV-1.

The inherent flexibility of the Env trimer, which transitions between conformations even when it is not bound to a cellular receptor, limits the options for structural analysis, because dynamic processes and metastable states cannot be captured by crystallization and cryo-electron microscopy. Our knowledge, therefore, rests on data obtained from analyses of Env proteins that have been artificially stabilized, such as engineered mutant Env proteins or complexes between Env and antibodies[2–4]. Two distinct conformations of the Env trimer — one closed, and one open and bound to the CD4 receptor — have been defined by structural analyses.

Structures known as SOSIP trimers comprise one type of engineered stabilized Env trimer that has been investigated for immunization, and in structural studies[3]. The capacity of antibodies to bind SOSIP trimers correlates with the extent of their neutralization breadth[3,5]. This indicates that these trimers represent a native, neutralization-relevant conformation of the Env protein, which makes them promising candidate immunogens for vaccines.

A technique called single-molecule fluorescence resonance energy transfer (smFRET) imaging provides information on the proximity of specifically labelled protein regions, which changes with the protein's conformational state. This technique does not allow visualization of the Env protein's overall structure, but tracks the opening and closing of the Env trimer. Three Env states can be distinguished before fusion of the virus with host cells: state 1, an unbound state that shows the highest degree of closeness; state 2, a slightly more open, intermediate state; and state 3, an open, CD4-bound state[6] (Fig. 1). Lu and colleagues used smFRET on stabilized Env variants investigated in structural analyses to understand which of the smFRET states matches the established models of Env-trimer structure. They also analysed the wild-type Env that is expressed on virus particles.

Through a series of technically challenging measurements, the authors show that the stabilized Env trimers predominantly adopt the intermediate state 2 conformation, and not the supposedly more closed state 1 adopted by wild-type Env. Four bnAbs isolated from cows vaccinated with a state 2 SOSIP Env immunogen[7] also targeted the state 2 conformation of Env. This finding suggests that antibodies induced by vaccination might preferentially recognize the conformational state of whichever Env immunogen is used in the vaccine. By contrast, and astonishingly, most HIV-1 bnAbs isolated from HIV-1-infected people that were assessed in this study targeted the state 1 conformation.

Given that the Env immunogens currently under investigation for vaccine development are in a state 2 conformation[2–4], these observations call scientists to action. Which of the unbound conformations of Env, state 1 or state 2, is the better target for neutralizing the virus? Which state triggers bnAb responses in a natural infection? And which state provides the best immunogen for vaccination? Defining the structure of the elusive state 1 and its functional properties is essential to answer

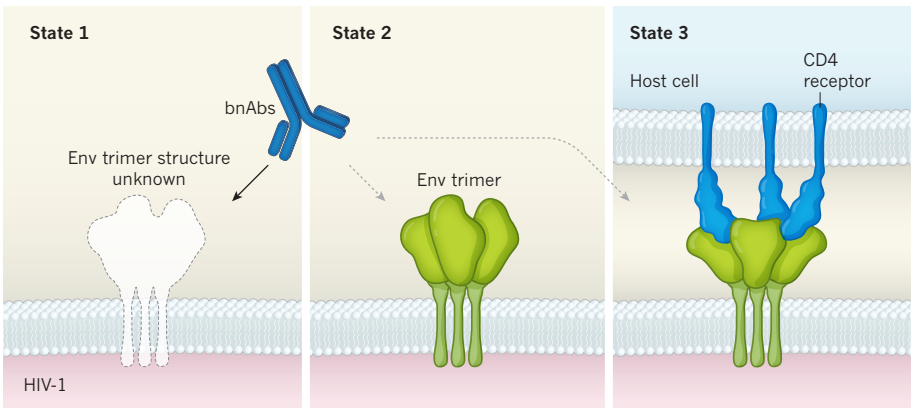

Figure 1 | Conformational states of the envelope (Env) glycoprotein of HIV-1. Env is a trimeric glycoprotein that has a closed conformation before the virus attaches itself to the cells it infects. On binding to CD4 receptors on host cells, Env is in an open conformational state, the structure of which is known (shown as state 3). Previous studies have also defined the structure of a closed conformational state of Env, and efforts to develop HIV-1 vaccines have been directed at this state. However, using single-molecule fluorescence resonance energy transfer (smFRET) imaging, Lu *et al.*[1] show that the closed conformational state defined by structural analysis is an intermediate, partially opened state (shown as state 2), and that structural information about the most-closed state (shown as state 1) is still missing. Broadly neutralizing antibodies (bnAbs, which neutralize various subtypes and variants of a virus) that were isolated from people with HIV-1 preferentially target state 1 of Env. This suggests that this conformational state might induce the production of bnAbs and is relevant for vaccine development.

these questions. Furthermore, researchers need to identify specific inhibitors for distinct Env states, because they will indicate which conformations are neutralization-relevant targets.

As intriguing as the observations by Lu and colleagues are, much remains to be done to understand the implications fully. A conformation that predominates on native virus particles might not be a good immunogen, despite its physiological relevance, if it cannot be engineered to be stable. Furthermore, any differences between states 1 and 2 might be too small to induce different bnAb responses. Only a head-to-head comparison of vaccines using immunogens based on both Env conformations will address this point. Although state 1 immunogens are not available, upcoming vaccine trials with state 2 immunogens will bring relevant data to this debate.

Defining the relative importance of the different conformational states of the Env trimer of HIV-1 before its fusion with host cells resembles the task Dutch artist M. C. Escher carved out for viewers in his 1938 optical illusion *Three Birds* (see go.nature.com/2upmu3f). Much like choosing the most dominant colour of bird in his fluttering flock, selecting the most relevant Env conformation among the transitioning pre-CD4-bound states of the trimer depends on the context in which they are viewed. Although the differences between states 1 and 2 might be subtle, defining these states structurally and functionally is essential to inform the HIV-vaccine field. We can only be sure that the bird in the hand is indeed worth two in the bush if we seek them all out. ■

Alexandra Trkola *is at the Institute of Medical Virology, University of Zurich, Zurich CH-8057, Switzerland.*
e-mail: trkola.alexandra@virology.uzh.ch

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CONDENSED-MATTER PHYSICS

Electrifying skyrmion bubbles

An electrical analogue of the magnetic-skyrmion bubble — a swirling arrangement of magnetic moments — has been unveiled in an artificially layered oxide material, raising prospects of new physics and applications. SEE LETTER P.368

PAVLO ZUBKO

Like a pesky cowlick that can't be tamed no matter how much you threaten it with a comb, tiny whorls of magnetic moments (spins), known as skyrmions and found in magnetic materials, can be extremely persistent, thanks to their specific topology[1]. And, just like hairdos, skyrmions and their various relatives come in many shapes and sizes, and with a mishmash of unusual names, such as hedgehogs, anti-hedgehogs and skyrmion bubbles. Skyrmions have been thoroughly studied since their experimental observation a decade ago[2,3] and promise denser and faster magnetic data-storage devices, but their electrical analogues have been elusive. Now, in a combined experimental and theoretical study on page 368, Das *et al.*[4] demonstrate that ordered arrays of polar-skyrmion bubbles — electrical cousins of magnetic-skyrmion bubbles — can be stabilized in artificially layered oxide materials.

Despite their fundamentally different physical origins, materials called ferroelectrics and their eponymous magnetic counterparts, ferromagnets, have many similarities. The defining property of a ferroelectric (or ferromagnet) is a spontaneous electrical polarization (or magnetization) that can be reversed by the application of an electric (or magnetic) field. This attribute makes both of these materials extremely useful for data storage, as well as for a multitude of other applications. However, unlike spins in magnets, which can often rotate with relative ease to give complex, swirling patterns, electric dipoles that arise from the relative displacements of positive and negative ions in a crystal cause a deformation of the crystal lattice, and must pay a hefty price in elastic energy to bend outside the ordered ranks.

Nevertheless, this difference hasn't stopped researchers from looking for patterns of rotating polarization in ferroelectrics, and one way to bend the rules is to go small[5–8]. When a ferroelectric is confined to the nanometre scale, it can be subject to large internal electric fields and stresses. These can strongly perturb the local polarization orientation and produce highly non-uniform distributions of electric dipoles, especially near surfaces or interfaces and domain walls, which are boundaries separating regions (domains)

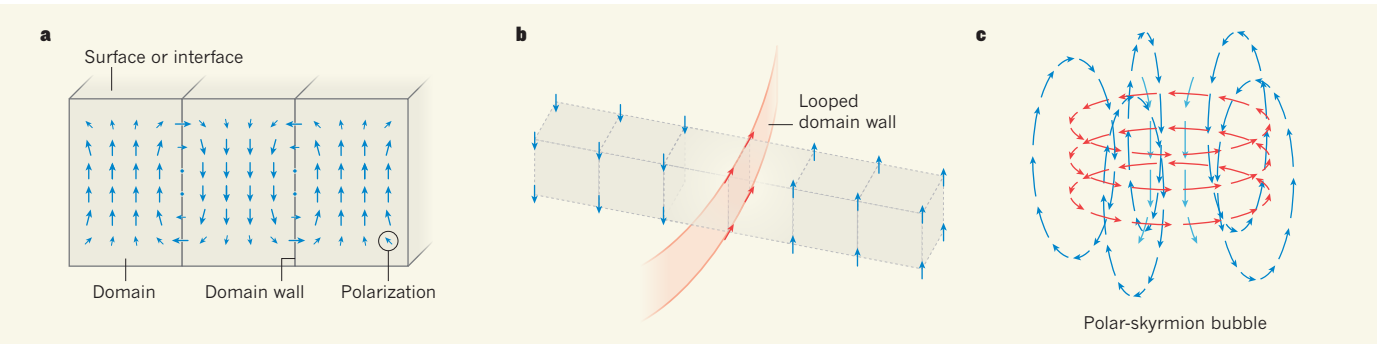

Figure 1 | The making of a polar-skyrmion bubble. **a**, When materials known as ferroelectrics are confined to the nanometre scale, they form tiny domains — regions of opposite electrical polarization. Near the top and bottom surfaces or interfaces of the material, the polarization can change magnitude and orientation, causing the polarization to rotate across domain walls. **b**, Ferroelectric domain walls can host polarization components perpendicular to those in the adjacent domains. Therefore, if such a domain wall is looped, it can form a ring of rotating polarization. **c**, Das *et al.*[4] report the observation of an exotic polarization pattern called a polar-skyrmion bubble. This pattern can be viewed as arising from nanometre-scale looped domain walls that combine the two types of polarization rotation in **a** and **b**.