as the energy of trapped charge carriers is stimulated to migrate to lanthanide ions. The resulting image can simply be recorded using a digital camera or smartphone.

The authors demonstrated the capabilities of their technique by using it to visualize the internal structures of curved circuit boards. They found that the resolution of the images can be increased if the detector is stretched when it is applied to the object under investigation. By embedding nanocrystals in a highly stretchable silicone, the authors achieved a resolution of about 25 micrometres. This is much higher than the resolution that can be achieved using conventional flat-panel detectors (typically in the order of 100 micrometres).

Several issues will need to be addressed before Xr-LEI can be translated into medical and industrial applications. For example, there is room for improvement in the X-ray sensitivity of the detectors, because only a small amount of nanoparticles (about 2% by weight) is incorporated into the silicone sheet. More fundamentally, work is also needed to understand exactly how Frenkel defects affect luminescence, but this presents challenges — for instance, identifying Frenkel defects formed by X-ray irradiation is difficult. Advanced techniques, such as time-resolved X-ray absorption spectroscopy and solid-state nuclear magnetic resonance spectroscopy, could be used to directly probe the change of positions of fluoride ions that lead to defect formation. Nevertheless, the insights presented by Ou *et al.* open up a promising avenue of research that might provide a new approach for non-invasive medical radiology and inspection of nanoelectronics.

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Ebola virus

Antibody highs and lows in survivors

Courtney Woolsey & Thomas W. Geisbert

For those fortunate enough to have survived a deadly disease, a vital question remains: how long does their hard-earned immunity last? Tracking of antibodies in Ebola survivors reveals a surprising pattern. **See p.468**

The West African Ebola epidemic in 2013–16 was unprecedented in its size, affecting more than 28,000 people. A more recent outbreak in the Democratic Republic of the Congo that ended last June added more than 3,000 cases to this total. The scale of these outbreaks is devastating, but there is nevertheless a large pool of survivors. This offers a welcome chance to study the stability of anti-Ebola antibodies — an indication of whether the body's immune system remembers the virus. Numerous groups have reported patients' antibody concentrations during the acute phase of Ebola disease and immediately after, but few studies have measured these levels long after recovery. On page 468, Adaken and colleagues[1] address this knowledge gap.

Antibodies are a crucial component of adaptive immunity — the arm of the immune system that responds in a specific (rather than a generalized) way to a foreign body. During a viral infection, the immune system scales up the production of antibodies that target part of a viral protein (an antigen). These either directly neutralize the virus by stopping it from entering a host cell, or destroy it indirectly by coating it, clumping viral particles together and generally doing everything possible to draw the attention of immune cells. When a person recovers from an infection, specific, long-lived plasma cells that reside in the bone marrow retain a memory of the virus, secreting numerous antibodies when re-exposed to the same antigen. These antibodies, called immunoglobulins, come in various forms — A, D, E, G and M — that have different activities and are found in different locations. IgG is the most common antibody in blood serum and extracellular fluids.

These are the basics of antibody activity, but different viruses have different effects on immunity. Vaccination or previous infection can provide lifelong antibody-mediated immunity to measles and mumps. But a seasonal influenza shot typically imparts immunity against a particular flu strain for only about a year. This discrepancy is attributed to factors that help to maintain antigen-specific plasma cells[2].

Little is known about the immunological memory of past Ebola infection. Previous outbreaks have been small and sporadic; the large size of the more recent epidemics therefore offers a unique opportunity. Adaken *et al.* tracked IgG in 115 Ebola survivors for up to 500 days after infection, making it the first study of its kind. They found a rapid decline in IgG levels after recovery. However, a substantial proportion of survivors experienced antibody resurgence approximately 200–300 days after recovery; the antibody levels then decayed again. This 'decay–stimulation–decay' pattern suggests new antigenic stimulation without overt disease or detectable levels of virus.

To enable them to make this discovery, the researchers isolated blood plasma at different time points from healthy survivors following the outbreak in Sierra Leone. They then used two approaches to detect antibodies: an *in vitro* test to measure levels of neutralizing antibodies (those that render the virus harmless); and a solid-phase assay to gauge the total amounts of antigen-binding antibodies, both neutralizing and non-neutralizing (those that signal the presence of the virus to other immune cells).

For the *in vitro* test, Adaken *et al.* created synthetic virus particles (pseudo-particles) consisting of a lentivirus that had been engineered to express the Ebola surface glycoprotein. They also engineered human cells to include a gene encoding luciferase, which generates a bioluminescent signal when cells are infected with the lentivirus. When an appropriate substrate is added and comes into contact with luciferase, an enzymatic reaction occurs, emitting light at levels that are directly proportional to the number of pseudo-particles that entered the cells. So, greater lentivirus infection means more light is emitted (Fig. 1a).

The authors placed the engineered cells in the wells of a test plate, along with survivor plasma and a fixed concentration of pseudo-particles, and then monitored light

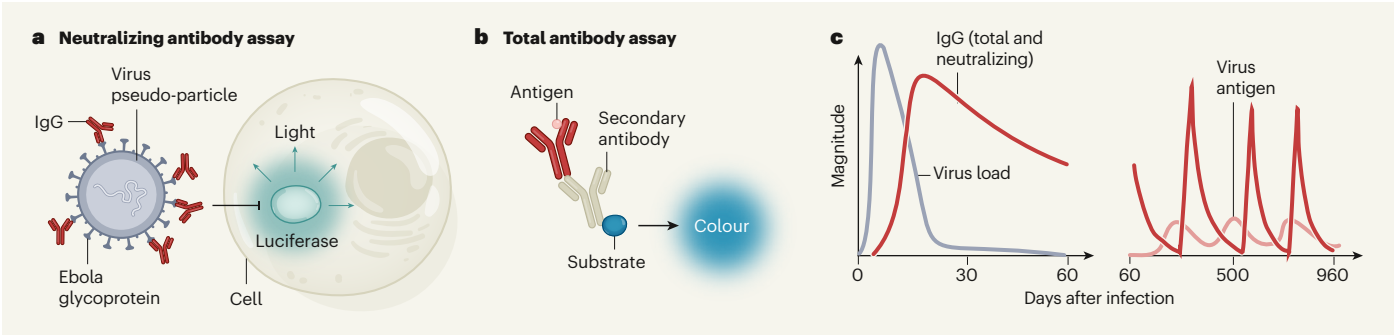

Figure 1 | Testing antibody levels in Ebola survivors. At different time points between 30 and 500 days after infection with Ebola, survivors donated blood. Adaken *et al.*[1] tested the levels of IgG antibodies against Ebola in the plasma from this blood — both a subset called neutralizing IgG antibodies and total IgG. **a**, For the neutralizing-antibody assay, the authors added blood plasma to test wells containing engineered cells that encode a light-emitting luciferase enzyme, together with synthetic viral particles (pseudo-particles) that express the Ebola surface glycoprotein and trigger luciferase expression. Neutralizing IgGs bind to the glycoprotein to prevent the pseudo-particle from activating luciferase. The more antibodies there are in plasma, the less light is generated. **b**, In the test for total antibody, a viral protein fragment called an antigen binds to a plasma IgG antibody. A secondary antibody binds to the IgG and is in turn bound by a substrate that generates colour when a substrate is added. **c**, These graphs, adapted from Fig. 3i of the paper, show IgG levels during an initial infection (the period over which there was a measurable virus load) and over the long term after recovery (from 60 days onwards). The two tests, combined with modelling approaches, indicate that antibody levels decay and rise at regular intervals after recovery, suggesting that virus antigen levels in the body are increasing and dipping over time.

emission. Wells containing higher levels of neutralizing antibodies emit less light, because the antibodies bind to the pseudo-particles, stopping them from entering cells. The main advantage of this approach — compared with experiments that investigate neutralization of live Ebola virus — is that it can be used outside a high-containment laboratory, making it easier and more accessible.

For the solid-phase assay, Adaken and colleagues coated wells with Ebola antigen, and again added survivor plasma, allowing plasma-derived anti-Ebola IgG to stick to the antigen. Next, they introduced 'secondary' antibodies that bind to the bound antibodies — these release colour when substrate is added and enzymatically processed, enabling the authors to measure the levels of plasma-derived antibodies (Fig. 1b).

These light- and colour-based assays showed that levels of neutralizing and total antibodies continued to fluctuate in survivor plasma. The authors combined these results with mathematical models to reveal the specific intervals over which antibody levels waxed and waned over time (Fig. 1c). Why do these fluctuations occur, given that Ebola virus was undetectable in the same plasma? The authors argue (although they did not address this possibility directly) that these cycles might follow periods of low-grade viral replication in tissues that are shielded from a full-blown immune response. These 'immunologically privileged sites' include the eyes, central nervous system and testes; tolerance to antigens here mitigates tissue inflammation and damage, but might allow the virus to endure. Lack of immunological control because of waning systemic antibody levels, combined with virus replication at these privileged sites, could seed recurrent small infections, provoking periodic antibody responses.

This mechanism is quite plausible. Many Ebola survivors continue to experience myriad symptoms — termed post-Ebola syndrome — including headaches, fatigue, vision disorders and musculoskeletal pain. In a study conducted by the Partnership for Research on Ebola Virus in Liberia (the PREVAIL III group), viral RNA was detected in the semen of 30% of survivors at least once on intermittent testing, and persisted for up to 40 months[3]. So this immunologically privileged site might indeed be a reservoir for recurrent infections, causing spikes in antibody production. (By contrast, the levels of antibodies directed at more-common viral and vaccine antigens — measles, mumps, rubella and varicella-zoster virus — remain remarkably stable in the absence of antigen stimulation[4].) These results support the idea that continuing low-level Ebola replication is the cause of surges in antibody levels.

However, in the same PREVAIL study, antibody levels did not correlate with symptoms of post-Ebola syndrome[3]. Other, less-well-understood factors that help to maintain levels of plasma cells and antibodies might therefore also be involved. One possibility is continuous proliferation of a type of immune cell called a memory B cell, which can mature into a plasma cell in response to certain signals. Another is the persistence of antigens on immune cells called follicular dendritic cells in organs such as the spleen; these might stimulate memory B cells, giving rise to plasma cells. A third possibility is that self-antigens (those embedded in the body's own cells) trigger 'cross-reactive' antibodies that mimic the virus antigen[5].

A major goal, then, will be to identify the root cause of antibody resurgence in Ebola survivors. But an immediate implication is that, because anti-Ebola antibody levels wane between bouts of resurgence, recurrent vaccine boosters in survivors (in addition to heightened surveillance) might be needed to maintain antibody titres and prevent viral reactivation and transmission. That said, some vaccines induce relatively stable antibody titres against Ebola[6], suggesting that the ebb and flow of antibody levels seen by Adaken and colleagues is a feature unique to naturally acquired immunity to the virus.

Antibodies to other viral infections also fall over time — including in people who have had mild COVID-19 (ref. 7). The mechanisms of antibody decay have not yet been defined for COVID-19, but recurrent vaccination might be necessary here, too, to achieve durable immunity, as for Ebola and seasonal flu. So, for many reasons, Adaken and colleagues' work is both topical and vital.

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