Titersafe For Easy Hemagglutination Inhibition Assays

Off-the-shelf reagent for Influenza Vaccine Development

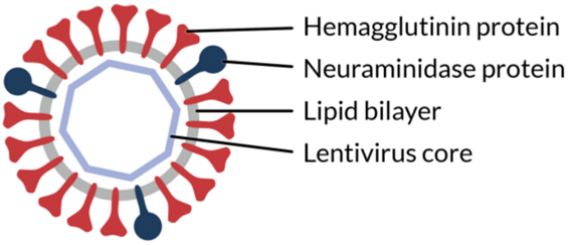
Get Ready for easier Influenza Assays

When you need to grow live influenza virus for hemagglutination inhibition (HAI) assays, it means moving other key tasks to the back burner. It also means exposing team members to occupational health risks.

With TiterSafe, you can get straight to work testing immune sera. The first product of its kind, TiterSafe is an off-the shelf reagent that can easily substitute for live virus in your existing agglutination assays.

Free from the chore of growing virus stocks, your team will have more time to focus on important science. Plus, because TiterSafe is non-replicative, your lab team will be safer.

TiterSafe HAI Reagent



Advantages

- ✔ Ready-to-use, quality controlled reagent
- ✔ Non-replicative & safe for BSL-2
- ✔ Easy to substitute in your existing assays
- ✔ Available for seasonal & custom strains

TiterSafe consists of membrane-wrapped particles with a lentivirus core. Its hemagglutinin (HA) and neuraminidase (NA) surface proteins have amino acid sequences identical to those from specific strains of influenza A or influenza B virus. TiterSafe has many advantages over live influenza virus in HAI assays.

What is TiterSafe?

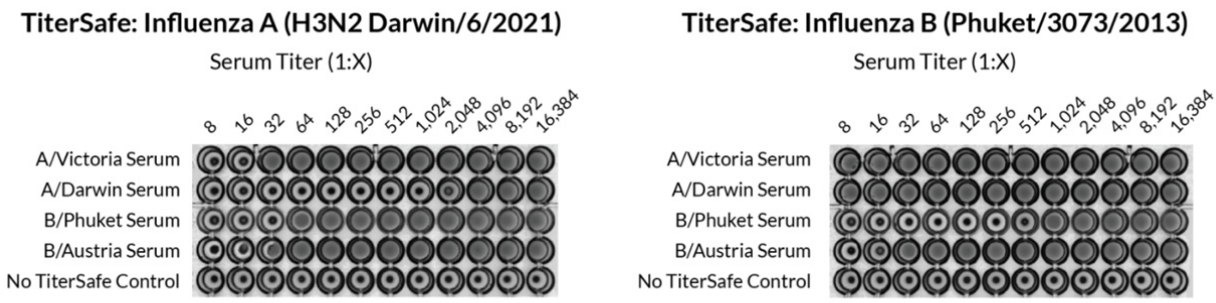
TiterSafe is a flexible reagent that can be used in a variety of influenza assays, including HA, HAI, and NA ELLA assays.

TiterSafe is based on our highly validated platform for building pseudotyped Reporter Virus Particles. TiterSafe is produced in human cells using transfected DNA encoding the influenza HA and NA proteins. Its HA and NA surface proteins have amino acid sequences identical to those in live virus.

TiterSafe's HA and NA surface proteins show the expected interactions and activities. In HAI assays with both sera and MAbs, TiterSafe shows the expected specificity for strain and lineage.

Because TiterSafe is not amplified by replication, the HA on the surface is the same on every particle and in every tube of reagent. Unlike with live virus, there is no sequence drift.

Due to its modular design, TiterSafe is flexible and highly customizable. By swapping out the DNA sequences, we can quickly and easily make TiterSafe product lines for new and custom influenza strains.



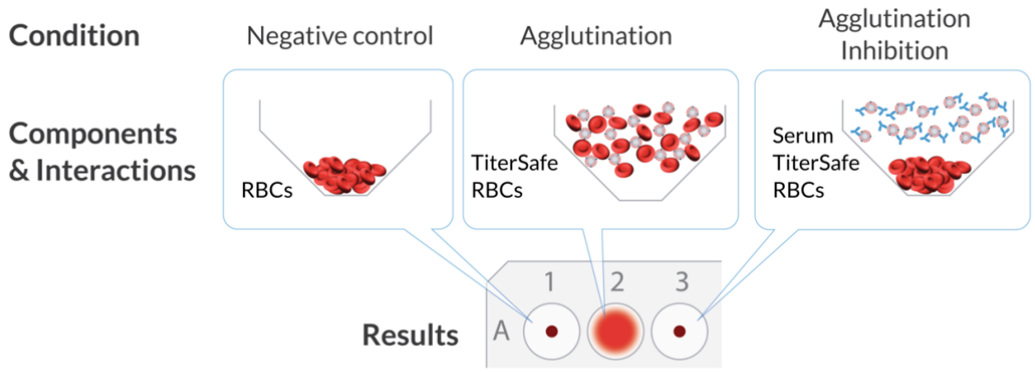
In sera and antibody screening, TiterSafe shows the expected specificity for strain and lineage.

How TiterSafe Replaces Live Influenza Virus!
In HA and HAI assays, TiterSafe performs like live influenza virus by cross-linking red blood cells (RBCs). This cross-linked matrix is visible to the eye as a broad red area spread across the sample well. In the absence of cross-linking, or agglutination, the RBCs form a concentrated spot at the bottom of the well.

As with live virus, you can use TiterSafe to detect agglutination-inhibiting antibodies in vaccine sera, and thus estimate the level of protection against influenza virus. When specific antibodies bind to TiterSafe surface proteins, they block agglutination, and the RBCs form a concentrated spot.

TiterSafe is an easy and safe substitute. It fits into conventional HAI workflows as a straight replacement for live influenza virus. It provides accurate HAI assay results, ensuring that your research remains reliable and valid.

Because TiterSafe has a much better safety profile than live influenza virus, it can be easier to work with. It is non-replicative and safe in a BSL-2 environment.



TiterSafe produces easy-to-read results in HAI assays.

Does TitreSafe contain Genetic Material?
No, there is no gene packaged inside the particles. This feature minimizes the risk of TiterSafe facilitating the integration of genetic material into any genome.

TitreSafe compared to Atteunated Influenza Virus!
TiterSafe can substitute in HAI assays not only for live influenza virus, but also for attenuated and inactivated influenza virus. Because some attenuated strains have modified HA sequences, TiterSafe may be even more representative of live virus than available attenuated technology. Unlike attenuated strains, TiterSafe eliminates the risk of infection.

How is TitreSafe different from RVPs?
TiterSafe is similar to Influenza Reporter Virus Particles (RVPs), but it lacks a reporter gene, and it is more concentrated.

Due to the similarities between the platforms we use to make the two products, we can provide matched reagents with the same surface proteins and overall structure: RVPs for neutralization assays, and TiterSafe for HAI assays.

Does TitreSafe have a Standardized Concentration?
While the concentration does vary somewhat between lots and product lines, TiterSafe always has sufficient titer to replace live virus in assays. Each tube of product meets the following minimum specifications:
The dilution for 1 HA unit is at least 1:256.
Each tube contains at least enough product for 1 HA plate (25uL) and 3 HAI plates using our recommended protocol.
Lot characteristics and recommended input volumes are included with product shipments.

Can TitreSafe be Used in ELLA assays?
Scientists at Integral Molecular have used TiterSafe in ELLA assays to characterize the activity of antibodies against influenza neuraminidase. Full support for this application including protocols and sample data is coming soon. For ELLA applications, we recommend reaching out about custom TiterSafe since for best ELLA results you may need a custom mismatch HA and NA.

ELLA Assays

Antibodies to neuraminidase (NA), the second most abundant surface protein on influenza virus, contribute toward protection against influenza. Traditional methods to measure NA inhibiting (NI) antibody titers are not practical for routine serology. The enzyme-linked lectin assay (ELLA), is a practical alternative method to measure NI titers that is performed in 96 well plates coated with a large glycoprotein substrate, fetuin. NA cleaves terminal sialic acids from fetuin, exposing the penultimate sugar, galactose. Peanut agglutinin (PNA) is a lectin with specificity for galactose and therefore the extent of desialylation can be quantified using a PNA-horseradish peroxidase conjugate, followed by addition of a chromogenic peroxidase substrate. The optical density that is measured is proportional to NA activity.

ELLA is an alternate assay to measure NI antibody titers first developed by Lambré et al. This assay quantifies enzyme activity by measuring the amount of the penultimate sugar of glycoproteins, galactose, which becomes exposed when sialic acid is released by NA. Since peanut-agglutinin (PNA) binds specifically to galactose, a PNA-horseradish peroxidase (HRPO) conjugate can be used to obtain a colorimetric read-out. The optical density that is measured is therefore proportional to the NA activity in the sample. NI titers are measured by determining the highest dilution of serum that inhibits at least 50% of the NA activity. This enzyme-linked lectin assay (ELLA) is performed in 96-well plates coated with fetuin, a highly glycosylated serum protein, as the substrate for NA.

The ELLA is suitable for measuring NI antibody titers in serum panels from preclinical and clinical influenza vaccine studies and can also be used to evaluate antigenic differences between the NAs of influenza viruses.

Anitha Florance or **Annie** as we fondly call her; is our Senior Application Scientist at KRISHGEN. She works on the product offerings from Integral Molecular products.

She has also been working over the past two years in the space of vaccines and biosimilar assays including neutralization assays, antigen-antibody PK and immunogenicity assays. Apart from this, she also coordinates the services for protein characterization, mapping and other offerings from us and our international partners.

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