One Minute Pitch

RNAscope ISH detects viral RNA, provides context, is highly sensitive and strand-specific; therefore delivers **where** the **actual virus** is and **what viral state it is in** (resting, replicating). Assay can be applied to **any sequenced virus,** already applied to >100 different viruses.

Pain Points & Our Solutions

Viral Location	**Context** provides viral localization which answers where virus entered, replicate, cause disease, shed
No Antibody	**Any Gene** of any sequenced virus, or for any gene in a virus.
Antibody Surrogate	**Direct** measurement of the actual virus not a protein that is produced as a host response
Low viremia	**High sensitivity** to detect single RNA molecules, proven for early infections and HIV/SIV reservoirs
Emerging Viruses	**Rapid** design and assay enable researchers to respond to emerging viral investigations
Viral State	**Strand-specific** probes discern viral stages; hybridize to sense (+) or anti-sense (-) strand to detect replication, resting stages
Viral Causality	**Localization & viral state** are needed to determine whether virus is just present or actually causing the disease
Uncommon species	**Any species** with a sequence. We have probes for viruses that infects raccoon, seal, fox
RNA/DNA	**RNAscope and/or DNAscope** to visualize viral states.

Solution for DNA & Retroviruses--DNAscope ISH

DNAscope ISH is a customer proven protocol. It can be used to detect viral DNA using a non-standard, modified protocol. It can be combined with RNA ISH so you see both viral RNA & DNA Defining HIV and SIV Reservoirs in Lymphoid Tissues.

Non-standard, can be cumbersome, recommend with caution

Key Tools / References

URLs https://acdbio.com/zika https://acdbio.com/HIV https://acdbio.com/infectiousdiseases

DNAscope Information

2 Spotlight Interviews Pesavento & Getu and Smits (Showpad)

Presentation Viral Pathogen Comprehensive PPT (Showpad)

Data Loads of publications & data images in PPT mentioned above

Who McCune Lab at UCSF for HIV, US CDC for Zika,

Research Goals

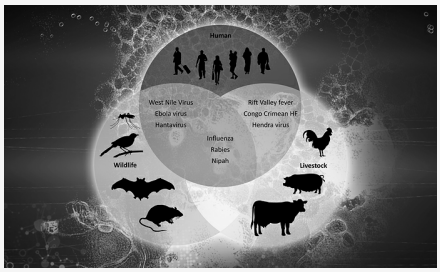
Viral pathogenesis how biological viruses cause diseases in their target hosts, Pathogenesis is a process in which an initial infection becomes a disease

Viral immunity correlates of immunity to infection and the development of novel approaches to the diagnosis, prevention and treatment

Viral emergence mechanisms that underpin host switching, examine same virus in multiple host species

Viral spread examine viral spread between livestock and wildlife. Examine resevoirs of infection and routes of transmission

Virus and Hosts



What is the viral type & viral group? Are there closely related species?

What is the host species and is there more than one? Is the parthenogenesis specific to the host?

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Qualifying Questions

Is your goal to locate the virus, find what state it is in or both?

> RNAscope ISH can provide location and viral state info

Which specific viral replication state do you want to detect? Attachment/Penetration/ Uncoating/Replication/Assembly/Release?

> For that specific viral state is it + /- stranded--probes designed complementary to that strand.

Is the antibody available for viral protein or is it a surrogate marker?

> RNAscope hybridize to the actual viral RNA and is strand specific, so it measures the actual virus in viral state.

Is low viremia a concern--early infection or viral reservoirs?

> Single molecule detection is proven with RNAscope. Several RNAscope HIV/SIV resevoir papers available.

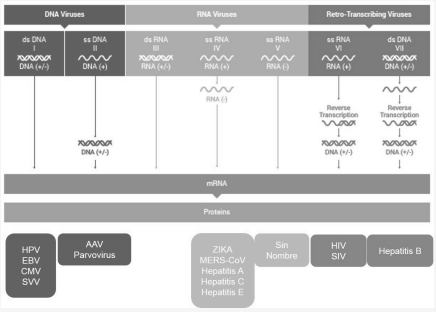
Do you need to link a viral stage to a disease? Do you want to detect a viral RNA and a disease marker?

> Multiplexing options available, so one probe can be design for the virus and another for the disease marker

Is host switching a concern?

> If the virus is the same in both hosts, then we can use one probe. But if the virus evolves from host to host, our probes are specific and be designed to the sequence

Background-- Compatible with all 7 Viral Groups



Publications using RNAscope ISH can be found for 6/7 viral groups

Background--Concepts & Vocabulary

Tropism	viral specificity for host tissue/cell (viral surface structures & host surface receptors)
Dynamics	speed of viral progression within host
Load/ Burden	quantity of virus in a given volume
Latency	ability of a pathogenic virus to lie dormant within a cell
Reservoir	cell type or anatomical site where (resting/latent) virus can hide, stay stable for future replication. May also be used to describe species that harbour the virus without causing disease.
Virions	virus while not inside an infected cell or in the process of infecting a cell
3 viral types	DNA, RNA (most common) & Retrovirus
7 viral groups	see chart
Sense (+) anti-sense virus	Sense virus hybridize with anti-sense probes, anti-sense virus hybridize with sense probes
Ambi-sense strand virus	single stranded genome with both +- strands requiring both sense & anti-sense probes
Host switching	species jumping, cross species transmission--a virus infects a new host species and may have changed at molecular level...new strain (SARS, Ebola, HIV)

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