Review of: "RNA in-situ hybridization for pathology-based diagnosis of feline infectious peritonitis (FIP): current diagnostics for FIP and comparison to the current gold standard"

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Potential competing interests: No potential competing interests to declare.

I liked this paper very much, but I wanted more details about your negative controls (which you rather vaguely described as “adequate”). In my limited experience of immunohistochemistry (IHC), the biggest problem we encountered was the “stickiness” of feline tissue, i.e., non-specific binding, causing false positive reactions. Consequently, my request is that you take a third sequential section of your sample blocks for running a negative control in your experiment in the exact same way as the ISH, but with a probe that would not detect feline coronavirus (FCoV): this would control for non-specific binding of the probe to the tissue. (Perhaps you did that, but didn’t include the photos?)

You have demonstrated greater sensitivity of ISH over IHC. If you intend to compare the specificity of ISH with IHC in a future publication, that would require further section(s) from the block(s), and a monoclonal antibody that does not detect FCoV, and you might also want to check whether your anti-mouse IgG binds non-specifically to that individual cat’s tissue. I cannot over-emphasise that it’s not enough to have a single negative control cat tissue for each batch of tests: you need to run negative controls on sections from the same block from each individual cat because of non-specific binding being a problem particular to each cat tissue.

I am hopeful that your technique will eliminate the problem of false positive FIP IHC reports by laboratories, and I look forward to seeing a row of four (as opposed to two) impressive photos from each cat in your next publication. And maybe a fifth photo with just traditional H & E staining for us old-fashioned pathologists? (I really liked your photos!)

I am tremendously excited by this publication and hope that it will usher in a whole new era of accurate FIP diagnosis. I especially hope that the authors will investigate using ISH on fine needle aspirates because biopsies are invasive and stressful for cats. That said, I don’t know if ISH would be quicker or have other advantages over quantitative feline coronavirus RT-PCR for use on fine needle aspirates?

Well done, Dr. Sweet and colleagues!