Reviewer's report

**Title:** Twelve years' detection of respiratory viruses by immunofluorescence in hospitalised children: impact of the introduction of a new respiratory picornavirus assay

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**Reviewer:** Hartwig Huemer

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The work of Sadeghi et al. describes their experience from the last 12 years with the use of direct immunofluorescence for detection of common respiratory viruses in the clinical setting.

I agree with the authors that in times of highly sensitive PCR methods, which may detect residuals of precedent infections for weeks or even months, a less sensitive test highly indicative for high viral loads in acute infection has still its value.

It is interesting that the rate of positivity of DFA (65%) in their hands nearly approached that of the Luminex test (78%) used in parallel.

I have 2 questions there: Which viruses have been missed in the DFA when compared to the Luminex? All the different ones, or were there any preferences?

Second, what was the rate of double infections in the Luminex? Was it also that low (less than 1%) than in the DFA? The role of double infections should be also addressed in the light of the assumptions of the authors that picorna (rhinovirus) may have an underestimated role in severe respiratory infection.

The point that HMPV is rather frequent, following Rhino and RSV and certainly Flu is well taken but I would not make a big point about the lack of a cyclic appearance. As HMPV is mainly a disease of smaller children, seasonal variations are expected especially in low birth populations where the relatively low numbers of susceptible children seem to lead to more extended cycles of infection with smaller outbreaks every 2-3 years, than in more densely populated areas with annual community outbreaks and more clear seasonal distribution. Frankly speaking the Swiss Alps may not be very representative in terms of HMPV transmission.

Figure 2 in the present form is very unclear and it is regrettable that the huge amount of information is rather hidden. Thus the seasonal cycles of viruses other than RSV and Flu are not visible at all due to the common scale for all of them. I would strongly suggest to split that grafic up in two parts and show the HMPV, PIV and Adeno separately with a smaller scale.

In summary I think that the work is sound and provides interesting data about an relatively old method, which although less sensitive than molecular methods has ist advantages in terms of specificity for acute respiratory infection.
Level of interest: An article of importance in its field

Quality of written English: Acceptable

Statistical review: No, the manuscript does not need to be seen by a statistician.

Declaration of competing interests:
There is no conflict of interest