

## Solutions for quantifying cytopathic effects after viral infection in vitro

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Individual cell-based virological assays require a maximum of experience an know-how. For the often time-consuming and costly experimental approaches, we introduce an outstanding technology for evaluating the results in a reliable and feasible way.

After different viral infections in vitro, the cytopathic effect (CPE) occurs, which changes the shape and appearance of the infected cells. In the early infection, only few cells show a CPE, which makes it different i.e. to estimate the success of an infection experiment. Also in later stages, quantification and qualification of CPE in stages 1-4 of CPE showing cells can be helpful to assess the right timepoint for different experimental approaches.

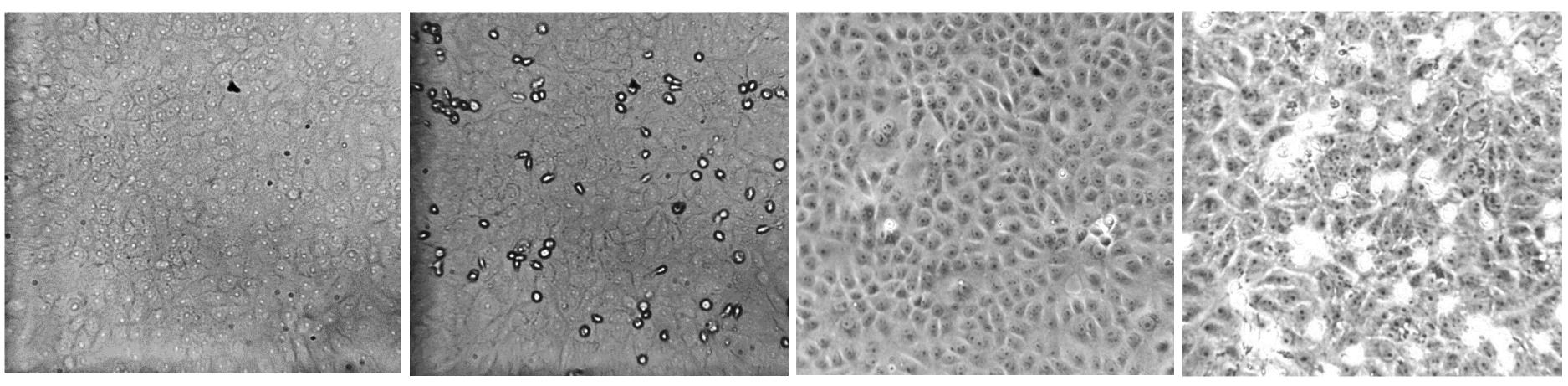


Fig.1 Examples of DMSO treated (B,D) or untreated cells (A,C), comparison of transmitted light (A,B) and phase contrast (C,D) in 10x magnification

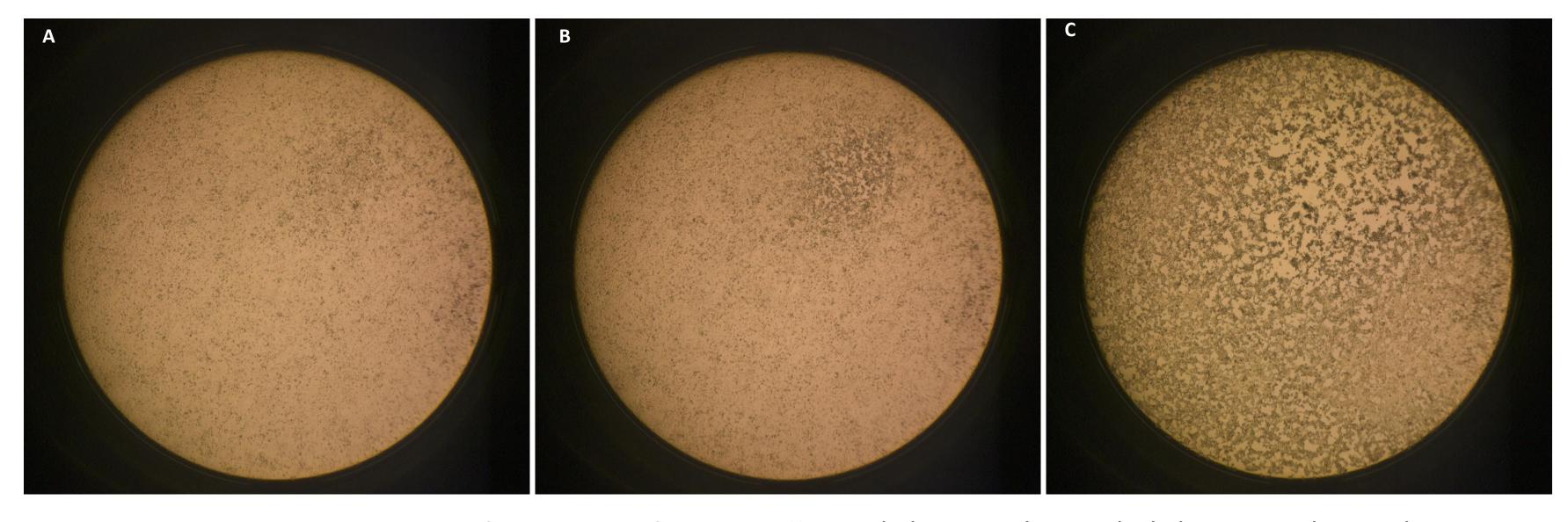
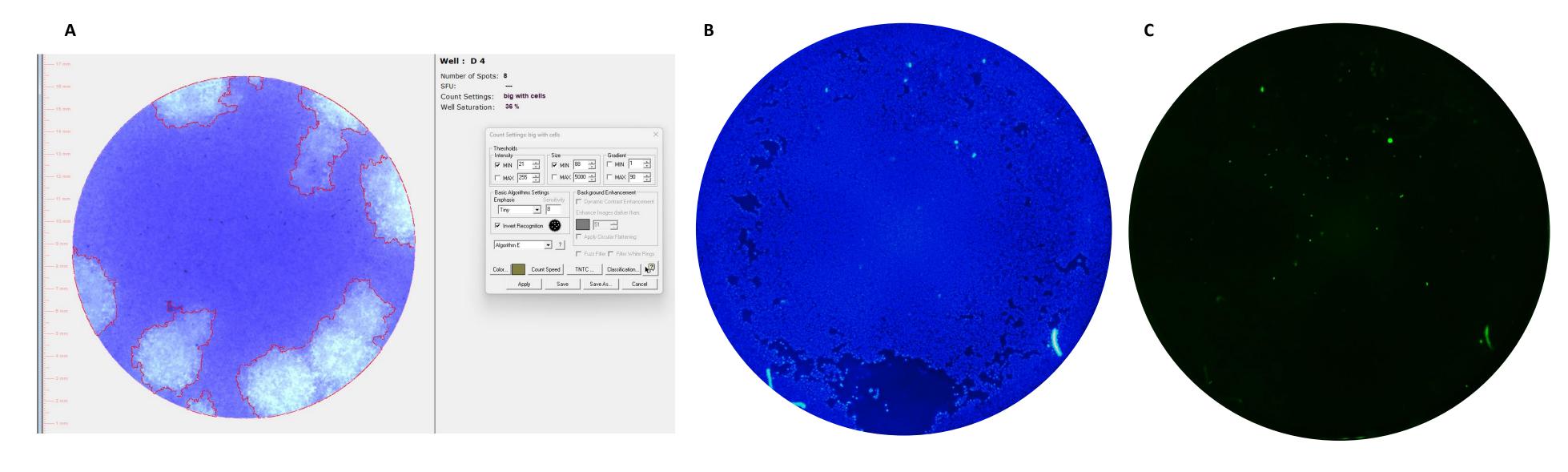


Fig.2 Dynamic measurement of vaccina infected cells on (A) day 9 (CPE 0), (B) day 10 (CPE 1) and (C) day 11 (CPE 4)



With the help of the AID multiSpot, a combination of an automated inverted microscope and a fluorescence EliSpot reader, this CPE can be monitored and quantified without cellular staining procedures and without interrupting any experimental approach directly in plates with liquid like medium.

Further virological assays like plaque assays are performed in different plate formats from 6-384 well plates, stained either with colorings like crystal violet or, mostly in 96-well format, with fluorescent dyes. The reader type for these (and many more) applications is the vSpot spectrum.

For visualization of these inventions, we treated cells with either DMSO (Fig.1) or infected them with virus preparations for plaque assays (Fig. 2) and measured the CPE over time or counted the resulted plaques. As the multiSpot is an automated microscope, one well can be completely scanned and therefore a whole well overview is possible in different magnifications from 4-fold to 20-fold. Comparison has also done for transmitted light or phase contrast (Fig. 1). The vSpot has the specialty to generate whole-well pictures from all plate formats due to its unique composition.

Fig.3 (A) viral plaque assay, well view, with counting dialogue example (red lines). (B) infected cells stained with DAPI and (C) GFP

Summarizing, this approach for the AID multiSpot enables to quantify and visualize easily the CPE in virus infected or compound treated cells and to get a complete overview of the whole wells that no important event is missed. This can be done in order to relate different treated cells showing a CPE and to make results between different experiments, concentrations or timepoints comparable.



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