NUF
FILTRATION
the new way of filtration

Possibilities of NUF filtration towards ToBRFV viral concentration

1. MATERIALS AND METHODS

The goal of the trial was to determine the presence of Tomato Brown Rugose Fruit Virus (ToBRFV) in water via (1) quantitative reverse transcription PCR (RT-qPCR, specific RT-qPCR for detecting ToBRFV with CaTa28 and CSP1325 primers and probe of the ISF protocol, 2020 conform the diagnostic standard EPPO PM7/146) analysis and (2) Agdia self-test (ImmunoStrip® for ToBRFV, Agdia, Elkhart, IN, USA) before and after concentration with the site NUF® pathogen detector.

The following schematic set-up was used (Fig. 1).

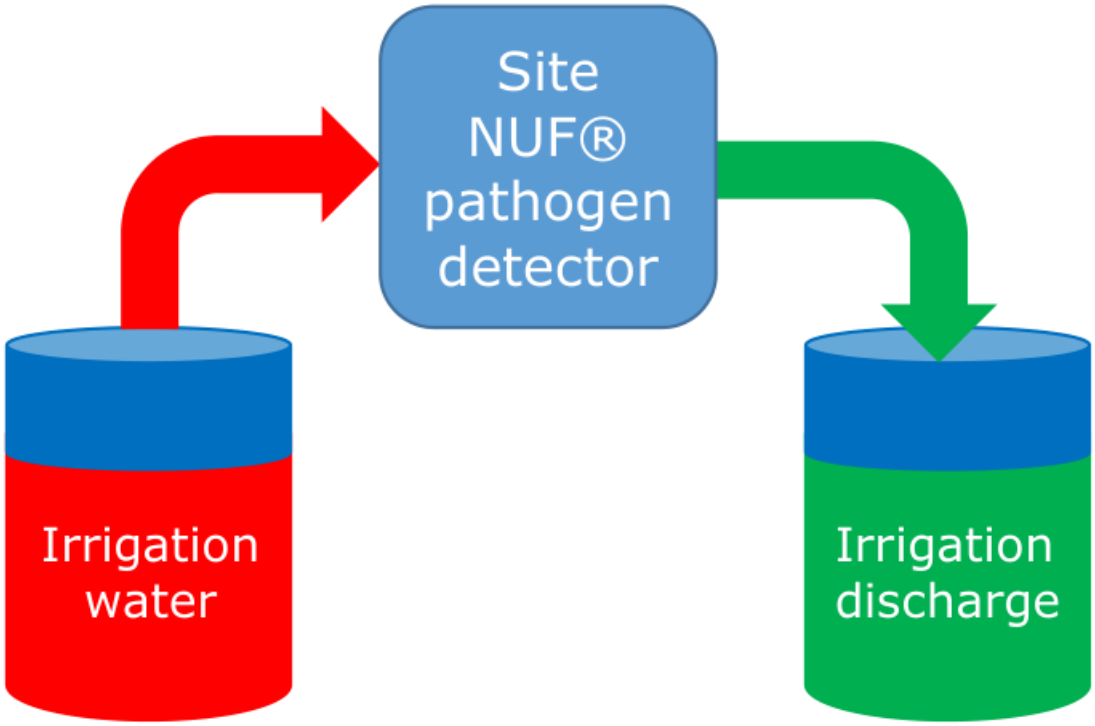

Figure 1: Schematic overview of the test trial set-up

The irrigation water was prepared by diluting one liter of ToBRFV contaminated irrigation water (positive control) in 600 liter of tap water. Afterwards 450 liter of the contaminated irrigation water was filtered over the site NUF® pathogen detector with an approximate flow rate of 95 liter/hour and collected in a new container. Subsequently, a backwash of the NUF membrane was done with 100 ml of sterile water, resulting in a 4,500x concentrated sample in comparison with the irrigation water. Samples were taken from the positive control, before filtration (irrigation

water after dilution), after filtration (irrigation discharge) and from the backwash of the NUF membrane. The (1) RT-qPCR analysis and (2) Agdia self-test were performed on all four water samples.

2. Results

Table 1 gives an overview of the results obtained for the (1) RT-qPCR analysis and (2) Agdia self-test. The pictures of the ImmunoStrips® of the Agdia self-test are shown in Figure 2.

Table 1: Overview of the qPCR and Agdia self-test results[1]

Sample	ToBRFV RT-qPCR results	Cq-value[a]	Result Agdia self-test[b]
Positive control	ToBRFV detected, high concentration	12.2	ToBRFV detected
Before filtration, Irrigation water after dilution	ToBRFV detected, medium concentration	21.4	ToBRFV not detected
After filtration, irrigation water discharge	ToBRFV not detected	34.6	ToBRFV not detected
Backwash	ToBRFV detected, high concentration	13.2	ToBRFV detected

[a] Threshold cycle of the qPCR analysis
[b] See Figure 2.

A high concentration of ToBRFV was detected in the positive control.

ToBRFV was not detected in the discharged irrigation water (after filtration), either with RT-qPCR analysis or with the Agdia self-test.

In the backwash water of the NUF membrane, ToBRFV was once again detected via RT-qPCR analysis as well as with the Agdia self-test.