

## **REPORT**

(15 February 2013)

### **Efficacy of Water Treatment with the AquaHort®-System against *Listeria innocua***

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**Client:**

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## Objective

To test the efficacy of the Aqua-Hort®-System against the bacterium *Listeria innocua* in a range of 0 to 4 ppm Cu at various exposure times (1 to 4 hours).

## Material and Methods

Aqua-Hort® Danmark ApS installed an Aqua-Hort®-unit at the experimental greenhouse of the Institute of Phytomedicin at the Geisenheim University. The unit has been put into operation and tested for its functional capability by Mr. De Lasson. He trained the personal involved into the project to handle and maintain the Aqua-Hort®-equipment.

400 L nutrient solution were prepared by 0.5 g/L of the complete fertiliser FERTY® 3 MEGA (ingredients see table 1). The solution ready for use had an electric conductivity of 1030 µS/cm and a pH of 6.4. Preparation took place one day in advance to achieve an adaptation to the ambient temperature of about 20 °C. The nutrient solution was pumped with about 1 m<sup>3</sup>/h through the Aqua-Hort®-unit. The Cu-concentrations were adjusted to 1, 2 and 4 ppm Cu according the various treatments and checked by using the "Kupfer-Test Aquaquant®" (range 0.3 – 5.0 mg/l; Merck KGaA, Darmstadt).

**table 1:** nutrient contents of the complete fertiliser FERTY® 3 MEGA (Planta Düngemittel GmbH, Regenstauf, Germany)

Plant Nutrients	Content (%)
nitrogen	18
potassium	12
phosphorus	18
calcium	2
boron	0.02
copper	0.04
iron *)	0.10
manganese	0.05
molybdenum	0.01
zinc	0.01
*) partially as chelate (EDDHA)	

After passage through the Aqua-Hort®-unit samples of 50 ml each were taken and contaminated with *Listeria innocua*. For this purpose from bacterial growth of 24 h plate cultures (TSA) a suspension with an OD<sub>590</sub> of 60 % transmission was prepared and 1 ml added to the 50 ml nutrient solution of each test tube. The samples were immediately transferred to the lab and stored there at room temperature. To realise the various exposure times three sub-samples each were plated onto TSA plates 1, 2 and 4 hours after the samples were contaminated (spiral plater; Meintrup DWS Laborgeräte GMBH, Löhden – Holte, Germany). At start and end of each treatment Cu-concentration (Kupfer-Test Aquaquant®; range 0.3 – 5.0 mg/l; Merck KGaA, Darmstadt), electric conductivity, pH and temperature of the nutrient solution were determined.

Each treatment was repeated four times. The viable counts (cfu/ml) were statistically analysed by ANOVA and significant differences to the control were determined for each exposure time by the Tukey-test (STATISTICA for Windows version 8.0).

## Results

After 1 hour exposure a significant reduction of the viable counts could be observed at 2 and 4 ppm Cu resulting in efficiency rates of about 40 and 60 % respectively (see table 2 and 3). At an exposure of 2 hours also with 1 ppm Cu the bacteria were significantly reduced (efficiency about 58 %) and with the two higher concentrations efficiency rates of 79 and 99.6 % were achieved. After 4 hours the efficiency rates were 97.57, 99.81 and 99.96 % for 1, 2 and 4 ppm Cu respectively.

**table 2:** Means of viable counts (cfu/ml) of *Listeria innocua* in a fertiliser solution after treatment at different Cu-concentrations and exposure times

treatment	exposure time		
	1 h	2h	4h
control	780,000 c	795,000 c	815,000 a
1.0 ppm Cu	668,333 bc	331,667 b	19,833 b
2.0 ppm Cu	491,667 ab	165,833 ab	1,583 b
4.0 ppm Cu	313,333 a	3,167 a	333 b

In each column values marked with the same letter are not significantly different; Tukey-test, p<0.05

**table 3:** Efficiency rates (per cent of control)

treatment	exposure time		
	1 h	2 h	4 h
1.0 ppm Cu	14.32	58.28	97.57
2.0 ppm Cu	36.97	79.14	99.81
4.0 ppm Cu	59.83	99.60	99.96

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(Prof. Dr. Walter Wohanka)