EFFICACY OF ELECTROLYSED ACID WATER AGAINST *PLASMOPARA VITICOLA* AND *ERYSIPHE NECATOR*

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Electrolyzed water is produced by electrolyzing purified tap water with the addition of a small quantity of saline solution (most usually NaCl or KCl). Anode and cathode are separated by a cation exchange membrane (fig. 1). At the anode, Cl_2, O_2 and H^+ are produced. Cl_2 then reacts with water and produces HClO and HCl. Small amount of H_2 and OH^- are produced at the cathode on the other side of the membrane. The cation exchange membrane between the electrodes allows movement of cations such as Na^+, but not anions, as OH^- (Tsuji et al., 1999). The water in the cell around the anode is acid, while the water in the cathode cell is basic. The electrolyzed acid water (EAW) has a pH lower than 2.7, mostly due to the HCl; and due to the HClO, which is oxidative, the redox potential (ORP) is over 1000 mV. The electrolyzed basic water (EBW) has a pH over 11 and a redox potential around -800mV.

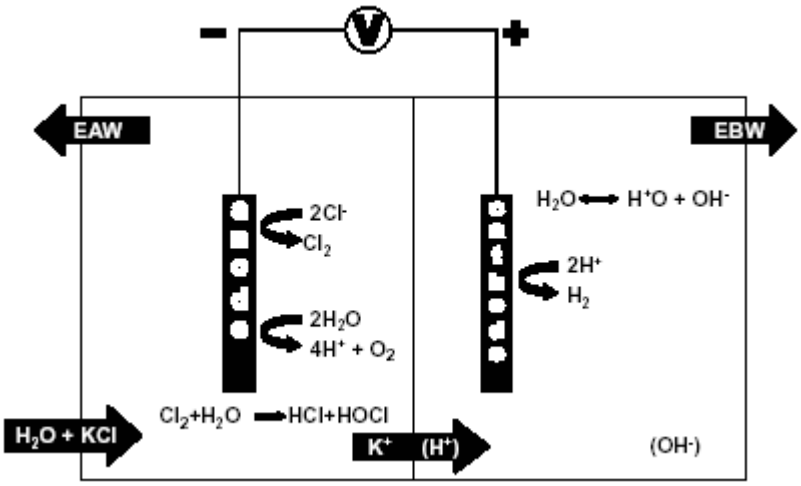

Fig. 1. The system for producing EAW and EWB

EAW has been used for over ten years as a disinfectant. It is widely used in Japanese hospitals for endoscope disinfection (Lee et al., 2004). The activity of EAW is based on its high redox potential (Tsuji et al., 1999), which causes damages on microorganism membrane (Kiura et al., 2001). The free-chlorine content of the EAW, enhances breaks and blebs on the membrane. Some studies on *Blumeria graminis* (Mueller et al., 2003) showed that EAW could have a possible uses in agriculture as a fungicide.

The aim of this research is to evaluate the potential use of EAW against the two main grapevine pathogens.
EAW was produced using Oxylyzer Ox 01 (CBCE, Italy). EAW was stabilized with the addition of EBW reaching pH 6±0.5 and an ORP of 650 mV. Deionised water was used as untreated control; copper hydroxide (Kocide 2000, Du Pont de Nemours) and wettable sulphur (Tiovit jet, Syngenta Crop Protection) were used as fungicide treated references respectively for downy and powdery mildew. The effect of experimental wetting agent X22 (Shin-Etzu, Japan) was tested, adding it (0.05 %) to water and to EAW.

Downy mildew

Trials were carried out on Pinot gris potted plants (5 plants/treatment). Plants were maintained under greenhouse controlled conditions (20° C), art ificially inoculated with a water suspension of *Plasmopara viticola* (Berk. et Curt) Berl. and De Toni sporangia (1x106 sporangia/ml) and overnight incubated at 20° C and 95% relative humid ity (RH). Since the efficacy of EAW last only few seconds, plants were treated with 100 ml of water, EAW with and without X22, 1, 3 or 7 hours after artificial inoculation, to see if EAW can interfere with the first stages of infection (stoma penetration and colonization). Copper and water controls were applied 6 hours before inoculation. Ten days after inoculation plants were incubated overnight at 20℃ and 95% RH to induce sporulation; afterwards severity (percentage of infected leaf surface) and incidence (percentage of infected leaves) were assessed.

Powdery mildew

Trials were carried out on Pinot gris potted plants (5 plants/treatment). Artificial inoculation was done shaking powdery mildew *Erysiphe necator* (syn.Uncinula necator) sporulating leaves. Plants were then incubated overnight at 20° C with high RH (80%). Daily treatments with water, EAW with and without X22 were applied for one week after inoculation. Sulphur was applied six hours before infection.

Statistical analyses of data were performed using the Statistica 6.0 software (Statsoft, Italy). Incidence and severity were respectively arcsin transformed to obtain constant variance, before analysis of variance. Duncan's test at $P<0.05$ was used to compare treatments. As shown in table 1, EAW was not able to significantly reduce P. viticola incidence, probably because of the high concentration of inoculum applied. A reduction in disease severity on leaves was seen only when X22 (in water and EAW) or copper were applied.

The results show that as soon as the pathogen has entered the stoma, X22 is not able to affect it.

Products	Time (hours)[1]	Dosage (ml/l)	Severity[2] (%)	Incidence[2] (%)
$Cu(OH)_2$	6 before	0.5[3]	16.5 a	68.7 a
X22	1 after	0.5	23.4 a	60.2 a
EAW+X 22	1 after	999.5+0.5	23.6 a	76.7 a
EAW	1 after	1000	40.5 ab	80.6 a
EAW+X 22	7 after	999.5+0.5	48.5 b	81.0 a
EAW+X 22	3 after	999.5+0.5	51.6 bc	75.3 a
X22	7 after	0.5	52.7 bc	65.4 a
EAW	3 after	1000	57.6 bc	77.0 a
X22	3 after	0.5	57.6 bc	77.1 a
Water	6 before	1000	61.6 bc	86.8 a
EAW	7 after	1000	65.3 c	78.1 a

Tab. 1. Results obtained against downy mildew with artificial infections under greenhouse controlled conditions. [1]Time from inoculation; [2]Means in the same column follower by different letters significantly differ (Duncan's test with P 0.05); [3] Expressed as g/l of copper .

Regarding powdery mildew (table 2) EAW and X22 were able to reduce disease severity and incidence, compared to water applied once before or daily after inoculation. Water daily applied was able to reduce the area of powdery mildew colonization on leaves (severity), but not the incidence.

Products	Time [1]	Dosage (ml/l)	Severity[2] (%)	Incidence[2] (%)
Sulphur	6 hours before	3[3]	2.3 a	44.0 a
Water	6 hours before	1000	13.5 d	100c
Water	1,2,3,4,5,6,7 days after	1000	6.1 c	100 c
X22	1,2,3,4,5,6,7 days after	0.5	3.6 ab	70.0 b
EAW	1,2,3,4,5,6,7 days after	1000	4.6 b	75.0 b
EAW+X22	1,2,3,4,5,6,7 days after	999.5+0.5	3.0 ab	53.3 ab

Tab. 2. Results obtained against powdery mildew with artificial infections under greenhouse controlled conditions [1]Time from inoculation; [2]Means in the same column follower by different letters significantly differ (Duncan's test with P 0.05); [3] Expressed as g/l of commercial product (Thiovit).

The results confirm what obtained by Mueller et al. (2003). EAW to be effective should be applied during inoculation. This is quite difficult with *P. viticola* whose infections occurs during rain, because EAW quickly loose its effect if applied on wet leaves. Practical applications could be very difficult since treatments one hour after inoculation are already ineffective against the disease.
On powdery mildew the efficacy of EAW is higher compared to downy mildew. This is probably due to the external growth of the fungus on leaves that can easily affected by EAW. The wetting agent X22 is fungitoxic by itself on the two pathogens, but also showed a high risk of phytotoxicity on grapevine.

The use of EAW could be an additional tool in IPM to reduce the fungicides against powdery mildew, but seems not practicable against downy mildew. Further studies will be necessary to define the right application timing on powdery mildew on grapevine.

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