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The Application of Acidic Electrolyzed Water to Grape Cultivation in the Southern Regions

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Abstract Experimental studies have shown that AEW (Acidic Electrolyzed Water) plays a strong role in killing grape virus, and it has instantaneous, broad-spectrum, efficient, safe and non-residual fungicidal properties. The results show that spraying AEW, and spraying alkaline electrolyzed water first and then AEW, can play a good role in controlling the virus damage of grape downy mildew, *Botrytis cinerea*, anthracnose, grape *Alternaria* spot, and anthracnose of grape, with foliar control effect of 93.5%, 96.2%, 93.20%, 100% and 87.9%, respectively, and ear control effect of 88%, 92%, 84%, 84% and 72%, respectively. AEW can replace pesticide, and can promote the average grape weight and sugar degree.

Key words AEW, Grape, Virus damage, Control effect

1 Introduction

Over the past decade, the table grape cultivation in the southern regions has become a characteristic fruit industry, and the varieties mainly include Kyoho, Xiahei, Gold Finger, Manicure Finger, Yongyou 1, Benifuji, Jumeigui, Shine-Muscat, Zuijinxiang, Rosario Bianco, Flame Seedless and Heibaladuo. The southern regions have rich heat resources for grape cultivation, but there is also a rainy and humid climate, easily causing frequent virus damage, which will increase the application of pesticides and pesticide residues, thereby having a direct impact on the economic benefit of farmers and industrial development. For example, the grape cultivation base in Jiangxi Xinyu National Agricultural Science and Technology Park is a local table grape cultivation base, which researches and develops AEW technology to control virus damage and achieve pollution-free cultivation, and it has achieved significant virus control effect.

2 AEW's virus damage prevention and control mechanism

AEW (Acidic Electrolyzed Water) is also known as the electrolyzed functional water^[1]. 0.1% sodium chloride (potassium) was added to purified water to be dissolved into the electrolyte solution having high conductivity. The electrolyzed reaction was generated in the electrolyzing device equipped with platinum electrodes, and it was electrolyzed into the sodium hypochlorite acid water with high oxidizing potential (ORP of +900- +1200mV). The pH value was 2-3, and the available chlorine content (ACC)

was 60-70 mg. L⁻¹. It also generated sodium hydroxide solution with strong reduction potential (ORP of -800 mV), and the pH value was 10 to 13. The principle of AEW in preventing and controlling virus damage and killing virus lies in the physicochemical properties of acid^[2-5]. The physical property is that it has high oxidation potential, and once it is in contact with bacteria and fungi or viruses, it acquires electrons from the biofilm with coerciveness and changes the normal potential and permeability of cell membrane, so that the permeability of the lipid membrane is damaged. It is like a hole through the cell membrane, leading to cellular content leakage. The chemical process is to rebuild a strong acid environment unsuitable for bacteria, thereby strongly inhibiting the bacterial development. Most of bacteria require pH of 3 or more, and the strong acid can have pH of 2.7 or less, so as to achieve bacteriostatic effects. The chlorine ion in hypochlorous acid is also a bactericide. After years of research, AEW has been gradually favored by people due to high efficiency, non-residue and non-pollution^[6-17].

3 Materials and experimental procedure

3.1 Materials, instruments and equipments The grape varieties for experiment include Xiahei, Gold Finger, Yongyou 1, Zuijinxiang, and Rosario Bianco, and the instruments can be shown in Table 1.

3.2 Experimental program

3.2.1 AEW preparation and determination of physicochemical properties The electrolyzed water was taken by the XY_150 device. The electrolysis voltage was 13V. The concentration of sodium chloride (NaCl) was 1% (prepared from pure water). pH value of the electrolyzed water was measured by the PHS-2C laboratory pH meter. The oxidation-reduction potential was provided by the equipment or measured by the ORP-501 oxidation-reduction potentiometer (PHSJ-4A). Based on GB/T601-2002, GB19106-2003, the available chlorine concentration (ACC) was measured by the

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RC-3F available chlorine detector. The physicochemical parameters of electrolyzed water can be shown in Table 2.

Table 1 Instruments and equipments used in the experiments

Instruments and equipments	Model number	Manufacturers
AEW production equipment	XY-150 electrical sterile water generator	Baoji Xinyuguang Mechanical and Electrical Co. , Ltd.
pH value tester	PHS-2C lab pH meter	Hefei Bridgesi Instrument and Equipment Co. , Ltd.
Available chlorine content measuring instrument	RC-3F available chlorine detector	Beijing Hangxuan Technology Development Co. , Ltd.
Balance and weighing machine	CLJ-0108 pallet scale	Beijing Zhonghui Tiancheng Technology Co. , Ltd.
Spraying equipment	F600 high-pressure sprayer	Heilongjiang Funong Machinery Co. , Ltd.

Table 2 Physicochemical parameters of electrolyzed water and tap water

Types	pH value	Available chlorine concentration//mg/L ⁻¹	ORP value//mV
Acidic electrolyzed water	2.4 ± 0.1	56.0 ± 2.0	1157.0 ± 3.0
Alkaline electrolyzed water	11.6 ± 0.1	0	-870.0 ± 3.0
Tap water	7.2 ± 0.1	0	680.0 ± 15.0

3.2.2 Experimental design and spraying method. The experiment was carried out from April to August 2015 in the grape cultivation base in Jiangxi Xinyu National Agricultural Science and Technology Park. Grape germinated in early April and was picked at the end of August. The vineyard uses the greenhouse shelter structure, and each greenhouse is 50 m long and 6 m wide. 25 plants were cultivated. There are five treatments: acidic electrolyzed water; alkaline electrolyzed water; alkaline electrolyzed water after acidic electrolyzed water; acidic electrolyzed water after alkaline electrolyzed water; bio-pesticides (control). There are five replications. The control experiment was conducted on five grape varieties. The hand-held high-pressure sprayer was used to spray electrolyzed water or biological pesticide on grape leaves and fruits from April 20 to August 26, and the spraying amount was 50 kg per mu. It was sprayed every 7-10 days, and when the grape virus damage was serious, it was sprayed every 3-5 days. It was sprayed with acidic electrolyzed water first and then alkaline electrolyzed water or with alkaline electrolyzed water first and then acidic electrolyzed water, with average interval of 30 minutes.

3.2.2 Control effect standard of grape virus damage. The grape leaf sprouted in early April, the leaf was infected with the Botrytis cinerea, downy mildew and brown spot in May 20, and the fruit

ear was naturally infected with anthracnose and grape Alternaria spot in June 10. Three points were randomly selected in each greenhouse before spraying electrolyzed water, in order to determine the infection area and viral species about the grape leaves. According to GB/T17980.30-2000 Field Efficacy Test Standard, the bactericide was used to prevent and control grape Botrytis cinerea, downy mildew, brown spot, anthracnose and grape Alternaria spot, and the disease index and control effect were calculated, respectively. Leaf virus damage grading standard is as follows: Level 0 (no scab); Level 1 (scab area accounting for less than 5% of total leaf area); Level 3 (scab area accounting for less than 6%-10% of total leaf area); Level 5 (scab area accounting for less than 11%-20% of total leaf area); Level 7 (scab area accounting for less than 21%-40% of total leaf area); Level 9 (scab area accounting for more than 40% of total leaf area). Fruit virus damage grading standard is as follows: Level 0 (no diseased fruit); Level 1 (diseased fruits accounting for less than 5% of total fruits); Level 3 (diseased fruits accounting for 6%-10% of total fruits); Level 5 (diseased fruits accounting for 11%-20% of total fruits); Level 7 (diseased fruits accounting for 21%-40% of total fruits); Level 9 (diseased fruits accounting for more than 40% of total fruits).

Foliar disease index =
$$\frac{\sum(\text{number of various levels of diseased leaves} \times \text{relative level value})}{\text{total number of leaves} \times 9} \times 100;$$

foliar control effect (%) =
$$\frac{\text{foliar disease index in the tap water control plot}-\text{foliar disease index in the prevention area}}{\text{foliar disease index in the tap water control plot}} \times 100;$$

fruit disease index =
$$\frac{\sum(\text{number of various levels of diseased fruits} \times \text{relative level value})}{\text{total number of fruits} \times 9} \times 100;$$

fruit control effect (%) =
$$\frac{\text{fruit disease index in the tap water control plot}-\text{fruit disease index in the prevention area}}{\text{fruit disease index in the tap water control plot}} \times 100.$$

3.2.3 Determination of main grape growth indicators. The grape plant height, rhizoma diameter, leaf area and leaf form were determined. Six grape plants were randomly selected at the beginning of experiment, and ruler and caliper were used to measure the plant height and rhizoma diameter weekly until the end of the experiment. Before spraying electrolyzed water each time, six ten-

der, middle and old leaves were randomly selected from six grape plants, respectively, and measured. The regression equation is as follows:
$$A = 0.64L^2 + 0.47W + 0.63W^2 - 0.62L \cdot W$$
where L is the maximum leaf length; W is the leaf width; A is the leaf area.

The mean of A was also calculated, and the chlorophyll content of leaf was measured at the same time.

3.2.4 Grape plant yield and quality measurement. After the grape picking period, the number of fruits per plant as well as the weight and number of fruits per string of grapes was calculated. The sugar meter was used to measure the sugar degree and alcoholic strength of the grape randomly selected.

3.3 Data analysis EXCEL 2010 was used for experimental data processing and statistical analysis.

4 Experimental results and analysis

4.1 Investigation of AEW prevention and control effect In the maturity period of grapes, the virus damage prevention and control effect was investigated once in the demonstration plot and control plot, respectively. 2 grape growing fields were randomly selected, and 5 sampling points were set in each field. 5 plants were investigated at each point, 20 leaves and 5 strings of ear were investigated per plant, a total of 25 plants, 500 leaves and 75 strings of ear. The number of leaves and fruit ears infected with downy mildew, *Botrytis cinerea*, anthracnose, grape *Alternaria* spot and anthrachose of grape was calculated, and leaf disease incidence, disease index and fruit ear disease incidence were also calculated. Throughout the demonstration process, the grape growth, growers' pesticide application frequency and rate, grape production and benefit were strictly recorded in the demonstrate plot and control plot, respectively.

4.2 AEW's grape virus prevention and control effect After AEW treatment of grape in the demonstration plot, the spread and

infection of some diseases (such as downy mildew, *Botrytis cinerea*, anthracnose, grape *Alternaria* spot, and anthrachose of grape) was greatly reduced or avoided, as shown in Fig. 1. The experiment effectively controlled the occurrence and spread of grape virus damage in the demonstration plot. According to the investigation in mid-July (grape maturity period) (Table 3), the leaf disease incidence, disease index and fruit ear disease incidence concerning grape downy mildew in the demonstration plot were 84.0%, 37.9% and 80% lower than in the control plot, respectively, with control effect of 94%, 93.5% and 88%, respectively; the leaf disease incidence, disease index, fruit ear disease incidence concerning *Botrytis cinerea* in the demonstration plot were 79.4%, 33.3% and 52% lower than in the control plot, respectively, with control effect of 95.4%, 96.2% and 92%, respectively; the leaf disease incidence, disease index, fruit ear disease incidence concerning grape anthracnose in the demonstration plot were 69.8%, 38.4% and 80% lower than in the control plot, respectively, with control effect of 97.6%, 93.2% and 84%, respectively; the leaf disease incidence, disease index, fruit ear disease incidence concerning grape *Alternaria* spot in the demonstration plot were 48%, 40.3% and 64% lower than in the control plot, respectively, with control effect of 100%, 100% and 84%, respectively; the leaf disease incidence, disease index, fruit ear disease incidence concerning anthrachose of grape were 89.8%, 39.5% and 56% lower than in the control plot, respectively, with control effect of 90.8%, 87.9% and 72%, respectively.

Table 3 AEW's grape virus prevention and control effect in Xinyu City during mid-July 2015

Treatment	Disease name	Number of diseased leaves	Disease incidence %	Diseased leaf control effect %	Disease index	Disease control effect %	Number of diseased fruits	Fruit ear disease incidence %	Diseased ear prevention and control effect// %
Demonstration plot	Downy mildew	30	6.0	94	1.6	93.5	9	12	88
	<i>Botrytis cinerea</i>	23	4.6	95.4	1.2	96.2	6	8	92
	Anthracnose	12	2.4	97.6	1.0	93.20	12	16	84
	grape <i>Alternaria</i> spot	0	0	100	0	100	12	16	84
	Anthrachose of grape	46	9.2	90.8	6.2	87.9	21	28	72
Control plot	Downy mildew	460	92	0	38.5	0	69	92	0
	<i>Botrytis cinerea</i>	420	84	0	34.5	0	45	60	0
	Anthracnose	360	72	0	39.4	0	72	96	0
	grape <i>Alternaria</i> spot	240	48	0	40.3	0	60	80	0
	Anthrachose of grape	450	90	0	45.7	0	63	84	0

4.3 Grape virus control effect under three treatments In order to study the electrical functional water control effect, three treatments (spraying acidic electrolyzed water; spraying alkaline electrolyzed water after spraying acidic electrolyzed water; spraying acidic electrolyzed water after spraying alkaline electrolyzed water) for comparative experiment, and it was compared with the treatment of spraying biopesticides. In view of the frequent downy mildew and *Botrytis cinerea* and serious disease characteristics during the grape cultivation in the southern regions, when the grape was infected with downy mildew or *Botrytis cinerea*, the bio-

pesticides and three electrolyzed water treatments were employed for study. It was found that there were significant differences in the final control effect of different treatments on grape downy mildew and *Botrytis cinerea* (June 23, 2015). As shown in Fig. 2, applying pesticide, applying alkaline electrolyzed water after applying acidic electrolyzed water, and applying acidic electrolyzed water, had good control effect, with disease index of 2.1, 2.8 and 2.2, respectively. The disease index of applying acidic electrolyzed water after applying alkaline electrolyzed water was 4.8. In this experiment, there was no significant difference in control

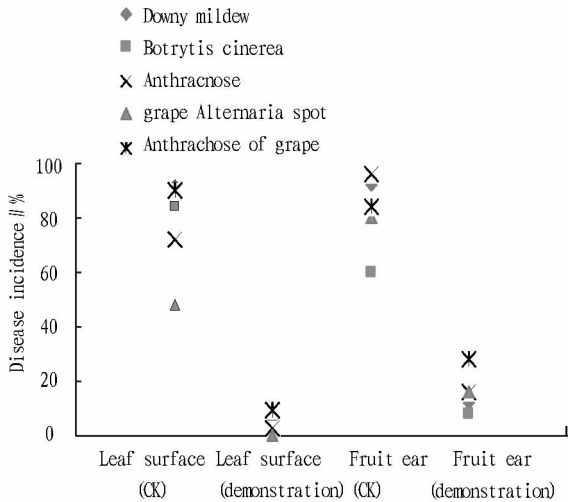


Fig.1 AEW's virus damage control effect

effect of grape downy mildew and Botrytis cinerea between bio-pesticides and applying acidic electrolyzed water or applying alkaline electrolyzed water after applying acidic electrolyzed water, while it was greatly different from applying alkaline electrolyzed water after applying acidic electrolyzed water. It can be found that applying acidic electrolyzed water, and applying alkaline electrolyzed water after applying acidic electrolyzed water, can have good control effect on grape downy mildew and Botrytis cinerea. In the prevention and control, there is a need to reduce the application of pesticides or avoid the use of pesticides, in order to reduce pesticide residues and environmental pollution. In addition, applying acidic electrolyzed water after alkaline electrolyzed water has ordinary control effect, and the reason needs to be further studied.

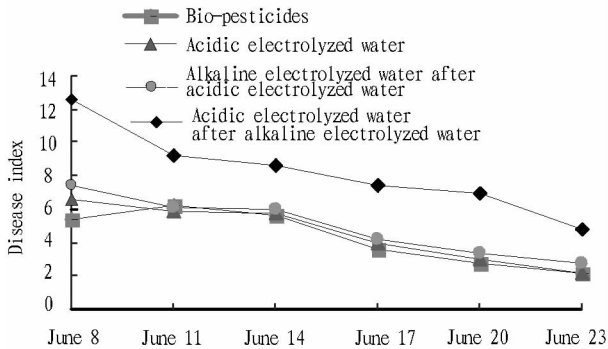


Fig.2 Control effect comparison of three treatments and bio-pesticides

4.3 Effect of electrolyzed water on grape weight and sugar degree For about every seven days, the Xiahei grape ripe during the same period is picked, and by sampling comparison, it is found that under the treatment of applying acidic electrolyzed water, the average weight or sugar degree of Xiahei grape are significantly higher than that of traditional varieties. As shown in Fig. 3, the average grape weight of Xiahei increases by 11.2%, and sugar degree increases by 9.6%, 1.8 points higher than that of normal varieties. It suggests that acidic electrolyzed water can pro-

mote grape growth and improve the quality of grape.

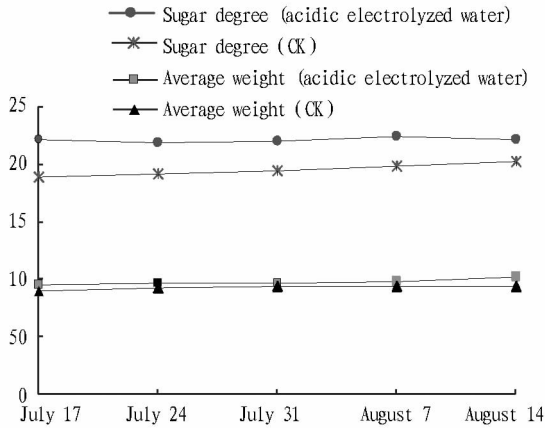


Fig.3 Effect of electrolyzed water on average grape weight and sugar degree

5 Conclusions and discussions

The experimental results show that applying acidic electrolyzed water, and applying alkaline electrolyzed water after applying acidic electrolyzed water have good effect in controlling some diseases such as grape downy mildew, Botrytis cinerea, anthracnose, grape Alternaria spot and anthrachose of grape (94% and 79.2%, respectively), and can replace pesticides. Acidic electrolyzed water can increase average grape weight and sugar degree, so acidic electrolyzed water can be safely applied in the greenhouse grape cultivation. The shortcoming in this experiment is that it lacks in-depth study of grape's Vc, soluble sugar, soluble protein content, and it will be addressed at the next stage. In this study, 150 L/h acidic electrolyzed water can be obtained from the water electrolysis device, and 1 h of water output can meet the application needs of five greenhouses (300 m²). The equipment is simple, and has low energy consumption and low electrolyte cost, with good promotion value. However, the electrolyzed water equipment has a high demand on water quality, and some areas with poor water quality need to be equipped with water softener. Studies have shown that acidic electrolyzed water plays a strong role in killing various types of microbes, and has good fungicidal properties^[18-19]. Compared with other chemical disinfectants, acidic electrolyzed water can be completely restored into non-toxic and non-residual ordinary water after sterilization, and it does not damage the ecological environment. The production cost of acidic electrolyzed water is low, and it is simple and easy to use, so it can be used as a kind of safe fungicide or disinfectant.

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sional knowledge, and resolve doubts. Classroom is the place where teachers get along with students frequently. In teaching, it is necessary to attentively analyze the learning state of students and observe students' class performance. If the students are absent-minded, dispirited, sleeping or play with the smartphone, the teachers should timely communicate with them and educate them. There is a need to encourage the students with mental burden, care for those students who need help, and tolerantly correct the error in students' studies. Teachers should use affection, experience and knowledge to set a good example to students and transfer positive social energy to students.

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(From page 87)

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