Application of Anatase TiO_2 Sol Derived from Peroxotitannic Acid in Crop Diseases Control and Growth Regulation

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Abstract

This paper reported the biological effects of TiO_2 sol on the control of crop diseases and growth regulation. The TiO_2 sol is a neutral, viscous aqueous colloid with size of 10-50 nm and 1.6% content of TiO_2, and was synthesized from peroxotitannic acid solution by Ichinose method. XRD result showed that the crystal form of TiO_2 nano-particles was anatase. Field experiment showed the TiO_2 sol formed a well-spread, transparent, and sticky layer of TiO_2 photocatalyst on the surface of plant, which could prevent plant from pathogens infection and improve photosynthetic performance. These results indicated that the nano-TiO_2 sol can be developed as a kind of environmental-friendly crop germicide and growth regulator.

Keywords: nanobiomaterial; TiO_2 sol; crop disease control; photosynthesis

INTRODUCTION

Synthetic germicides have been widely used to control crop diseases. However, they might cause problems such as environmental pollution and food safety issues. It has consequently become necessary to develop alternative methods for the control of crop diseases. The environmental friendly TiO_2 photo-catalyst as nanobiomaterials has shown potential for agricultural application because of its photocatalytic disinfection and photobiological effects[1-3].

Our group recently conducted experimental researches of biological effects of TiO_2 nano-particles on crop production. Preliminary results showed that TiO_2 photocatalyst had significant effects on preventing crop fungal/bacteria diseases and promoting photosynthesis [4-6]. But the suspension of TiO_2 nano-particles has low adhesion to plant leaves and therefore is difficult to fully fulfill their biological effects. This paper reported the biological effects of TiO_2 sol on crop production. Such TiO_2 sol was prepared from peroxotitannic acid solution (PAS TiO_2 sol) by Ichinose method [7-8] and is a neutral, viscous aqueous colloid of TiO_2

1 Materials and Methods

1.1 Preparation and Characterization of PAS TiO_2 sol

PAS TiO_2 sol was prepared from peroxotitannic acid according to the method described by Ichinose [1-3] using $TiCl_4$ and H_2O_2 as precursors. The TiO_2 sol was characterized by XRD (D/max-ⅢA, Japan) and TEM (JEOL-100CXII, Japan).

1.2 Evaluation of Bactericidal Effect

TiO_2-film coated glass slides (100 mm × 50 mm) used for bactericidal experiments were prepared by dipping with PAS TiO_2 sol and then heated at 160℃ for 3 h.

The photocatalytic bactericidal effect of PAS TiO_2 were evaluated by inoculating aqueous suspension of two kinds of pathogenic bacteria (*P. s.* pv. *lachrymans* and *X. vesicatoria*) on the surface of TiO_2 coated glass slides, respectively, followed by UV irradiation of black light (40W) for 12 h from the distance of 20 cm underneath.

1.3 Field Experiment

Cucumber (*Cucumis sativus L.*) was chosen as the experimental crop and cultivated in greenhouse according to normal production processes. At the 7 - 8 leaf-stage, cucumber leaves in treated blocks were sprayed with PAS TiO_2 sol using a manometric sprayer. Control experiments were conducted by spraying water in control blocks. Each treatment set three blocks with square of 2.7 m × 2.0 m in random distribution, and each block with 40 plants. Through experiment, each block was treated successively three times by spraying at an interval of 7 days.

1.4 Investigation of Effects on Diseases Control

The investigation of diseases control effects were performed by randomly selecting 20 seedlings in each block according to *GB/T* 17980.26-2000 [9]. .

1.5 Evaluation of Photosythetic Effect

Photosynthetic rate of cucumber leaves were measured by a portable photosynthesis system (Li-6400). The content of chlorophyll of leaves was analyzed by UV-VIS spectrophotometer (UV-2250, Shimatsu).

1.6 Statistical Analysis

Statistical analysis was done using SAS ver. 6.12 learn (SAS Systems, Cary, NC, USA). For evaluation the bactericidal properties of PAS TiO_2 sol, results were shown in the manner of ($\overline{x} \pm SE$). Significance of differences between control and treated samples was determined by analysis of variance followed by *t*-test at the 0.01 level of significance. For field experiment, results were analyzed statistically by analysis of variance (ANOVA), the least standard deviation (LSD) test was applied to make a comparison between treatments at the 0.05 level of significance.

2 RESULTS

2.1 Characteristics of PAS TiO_2 Sol

The characteristics of PAS TiO_2 sol were shown in Table 1.

TiO_2 (%)	Cu^{2+} (%)	H_2O (%)	Color	pH	Particle size (nm)
1.6	0.016	98.4	yellow	7.8	52.9

Table 1:Characteristics of PAS TiO_2 sol

As can be seen in Fig.1, the spindle-shaped PAS TiO_2 had an average particles size of 50nm. XRD patterns (Fig.2) confirmed that the crystal phase of TiO_2 was anatase. Therefore, PAS TiO_2 sol synthesized by Ichinose method in this study is a neutral, viscous aqueous colloid of TiO_2 with of 50nm and in the anatase crystal phase.

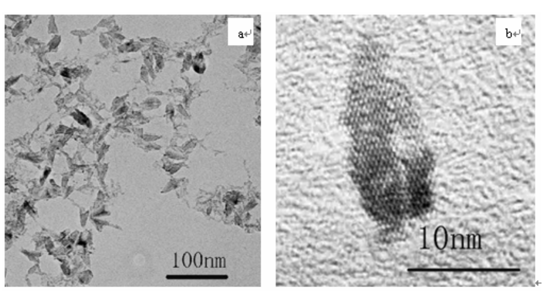

Figure 1 TEM images of TiO_2 sol

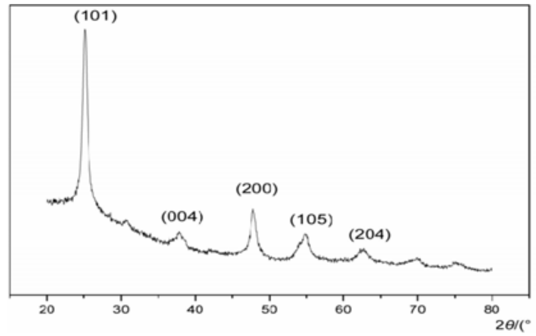

Figure 2 XRD patterns of TiO_2 sol

2.2 Bactericidal Effect of PAS TiO_2 Sol

It was observed that PAS TiO_2 sol in experiments had a perfect adhesive and film forming ability and could form continuous and stable films on the glass surface.

The photocatalytic bactericidal effects of PAS TiO_2 on pathogenic bacteria (*P. s.* pv. *lachrymans* and *X. vesicatoria*) were determined by colony counting 36 h after incubation. The result was shown in Table 2. More than 99% of bacterial cells on the surface of TiO_2-coated glass lost their viabilities after being illuminated for 12 h. The relative bactericidal rate of nano-TiO_2 sol to *P. s. pv. lachrymans* and *X. vesicatoria* was 99.9% and 100%, respectively.

pathogens	Treatments	Survival (CFU)	bactericidal rate（%）
P. s. pv. lachrymans	Control	1944 ±45.6A	-
	TiO_2-treated	1.6 ±0.5B	99.9
X. vesicatoria	Control	1393.7±37A	-
	TiO_2-treated	0B	100

Table2: Bactericidal rate of PAS TiO_2 sol

2.3 Diseases Control Effects of PAS TiO_2 Sol

PAS TiO_2 sol owned a perfect film-forming ability and powerful bactericidal ability on plant pathogenic bacteria. The field experiment further showed that spraying PAS TiO_2 sol on cucumber may form a well-spread, transparent and sticky TiO_2 photocatalyst layer on the surface of leaves, which could prevent plant from pathogens infection by photocatalytic disinfection.

The effects of PAS TiO_2 sol on cucumber diseases *P. s.* pv. *lachrymans* and *P. cubensis* were investigated in field cultivation experiment and the results were shown in Table3 and Table 4. The lesion areas, disease incidences and disease indexes of cucumber plant in TiO_2-treated blocks were significant less than that of control blocks

(P=0.05). The control efficiency of PAS TiO_2 sol to *P. s. pv. lachrymans* and *P. cubensis* is 68.6% and 90.6%, respectively. This indicated that PAS TiO_2 sol as a kind of germicides can effectively restrict the invasion of bacteria and fungal pathogens to crops.

Plant diseases	Treatment	Lesion areas		
		7^{th} leaves	8^{th} leaves	9^{th} leaves
P. s. pv. lachrymans	Control	13.3 a	18.0 a	12.1 a
	TiO_2-treated	3.9 b	6.5 b	5.6 b
P. cubensis	Control	18.8 a	26.43 a	33.5 a
	TiO_2-treated	4.5b	6.37 b	8.5 b

Table 3: Effects of PAS TiO_2 sol on lesion areas of cucumber diseases

Plant diseases	Treatment	Disease incidences	Disease indexes	Control efficiency
P. s. pv. *lachrymans*	Control	68.3 a	14.5 a	
	TiO_2-treated	42.7 b	6.3 b	68.6%
P. cubensis	Control	76.2 a	39.2 a	
	TiO_2-treated	26.8 b	7.8 b	90.6%

Table 4: Effects of PAS TiO_2 sol on preventing cucumber diseases

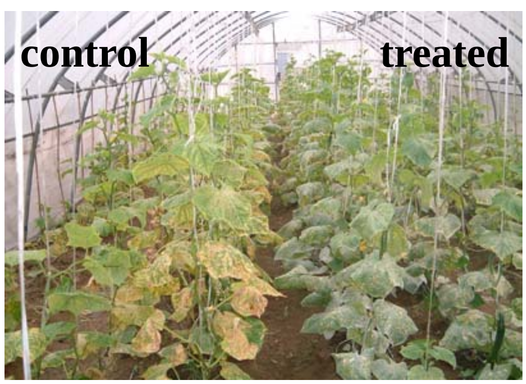

Figure 3 Field test of TiO_2 sol for the control of *Pseudoperonospora cubensis*

2.4 Effects of PAS TiO_2 Sol on Plant Photosynthesis

As shown in Fig.3, leaves sprayed with 0.8% PAS TiO_2 sol showed the best effects on promoting photosynthesis and caused about 30% increase in net photosynthetic rate. These experimental results indicated that spraying PAS TiO_2 sol on surface of cucumber leaves improved significantly photosynthesis.

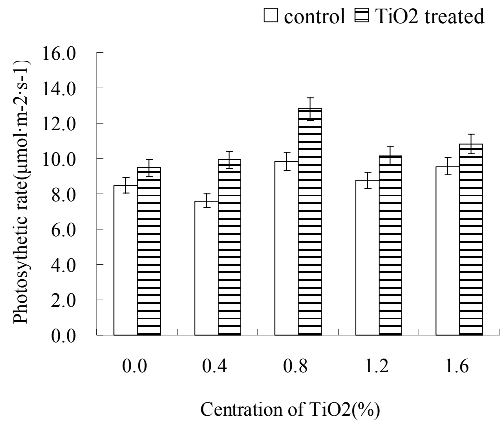

Figure 4 Effect of concentration of PAS TiO_2 sol on photosynthesis of cucumber

Fig. 5 indicted that net photosynthetic rate begin to increase significantly only 1 hour after PAS TiO_2 sol sprayed on the surface of leaves. But 1 hour later, the increase of net photosynthetic rate caused by PAS TiO_2 sol begin to decrease gradually. The physiological mechanism of TiO_2 in improving photosynthesis is still need to be studied in detail.

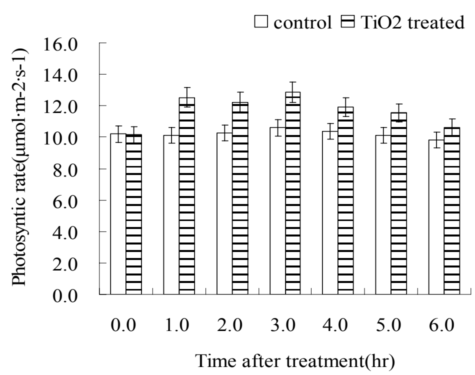

Figure 5 Changes of photosynthetic rate as function of time after PAS TiO_2 sol treatment

As was shown in Table 5, the photosynthetic pigment content of chlorophyll a, chlorophyll b in the leaves cucumber was also increased significantly after PAS TiO_2 sol treatment.

Treatments	Chlorophyll a	Chlorophyll b	Chlorophyll
Control	1.1761	0.4094	1.5855
TiO_2 sol	1.7263**	0.5363 *	2.2626**

Table 5: Effect of TiO_2 sol treatment on content of chlorophyll（** significant at 0.05 level, ** significant at 0.01 level*）

3 CONCLUSIONS

This study confirmed that PAS TiO_2 sol owns following biological effects:

(1) forming a successive, adhesive and transparent film on surfaces of leaves which causes photocatalystic disinfection and photobiological effects.

(2) possessing powerfully bactericidal effects to plant pathogens.

(3) blocking and inhibiting bacteria and fungal diseases of crops .

(4) promoting photosynthesis and chlorophyll synthesization.

Therefore, PAS TiO_2 sol is prosperous as a kind of environment-friendly germicide and regulator in controlling plant bacterial / fungal diseases and promoting plant growth.

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REFERENCES

[1] Yao KS, Cheng TC, Li SJ, Surface&Coatings Technology, 203 (5-7), 922-924, 2008.

[2] Eremia SAV, Chevalier-Lucia D, Radu GL, TALANTA, 77 (2), 858-862, 2008.

[3] Yao KS, Wang DY, Chang CY, Journal of Nanoscience and Nanotechnology ,8 (5) , 2699-2702, 2008 .

[4] Li Lingling, Cui Haixin; Zhang Ping. Transactions of the Chinese society of agricultural engineering, 24(8), 223-226, 2008.

[5] Zhang Ping, Cui Haixin, Li Lingling. Journal of Inorgannic Materials , 23(1),55-60, ,2008.

[6] Zhang Ping, Cui Haixin, Song Na, Li Lingling. Transactions of the Chinese society of agricultural engineering, 22(12), 13-16, 2006.

[7] Ichinose H., Katsuki H, J Ceram Soc Jpn, 106(3):344-347,1998

[8] Ichinose H., Kawahara A., Katsuki H., Journal of Ceramic Society of Japan, 104(10):914-917, 1996.

[9] GB / T 17980. 26—2000, Pesticide－guidelines for the field efficacy trials (I)—fungicides against downy mildew of cucumber [S].

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