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微生态制剂在葡萄上的促生防病效果

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摘　要：【目的】明确微生态制剂绿康威和绿地康对葡萄的田间促生防病效果，为实现葡萄的安全高效生产提供依据。**【方法】**在包头市以'巨峰'和'寒香蜜'葡萄为材料，比较微生态制剂和杀菌剂单独或交替使用的5个处理在生长量、产量和果实品质、对霜霉病的防效上的差别。**【结果】**微生态制剂的使用能显著促进'巨峰'和'寒香蜜'茎粗、茎长的生长，提高叶片叶绿素含量，同时增加产量和百粒质量，而对叶长、叶宽的影响不稳定，对可溶性固形物含量无影响。生长季在地上部和地下部同时使用绿康威和绿地康7次，对'巨峰'葡萄的霜霉病防效最好，可达75.68%；而田间常规用药防效仅为38.74%，单独地下部施用绿地康的防效为57.66%。**【结论】**地上部和地下部同时使用微生态制剂对葡萄促生长和霜霉病绿色防控有良好效果。

关键词：葡萄；微生态制剂；生长量；葡萄霜霉病；产量

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Grape growth enhancement and disease promotion by microecologics agents in Baotou

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Abstract:【Objective】The study was carried out to clarify the effect of microecologics Lvkangwei and Lvdikang on the growth and disease incidence of field grown grape, and explore the best application plan, so as to produce safe grapes efficiently. Lvkangwei and Lvdikang are microecologics in different dosage forms mainly composed of three strains of bacillus. They are used to improve the micro-environment of plants by applying to the above or the below ground.【Methods】Rain-sheltered 'Suffolk Red seedless' and open field 'Kyoho' grown in Baotou were used as materials for the experiment from May to September 2018, and five treatments were performed in a randomized block design. Treatment 1 was control, where water was sprayed evenly on the front and back sides of the leaves. Treatment 2 involved conventional chemical control scheme with no chemicals used from May 11 to June 2 but later with sprays of 40% chlorothalonil diluted 600 times, 25% metalaxyl · propamocarb hydrochloride diluted 700 times , 40% chlorothalonil diluted 600 times, or 70% Propamocarb diluted 1 000 times. Treatment 3 involved root irrigation of Lvdikang at 500 dilution times and foliage spray of Lvkangwei diluted 700 times for seven times. Treatment 4 was root irrigation of Lvdikang diluted 500 times and foliage spray of Lvkangwei diluted 700 times for four times, followed by conventional chemical application for the last three times. Treatment 5 was root irrigation of Lvdikang diluted 500 times alone for seven times. The interval between applications in all treatments was about 15 days. The diameters and lengths

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of the upper, middle and lower current season shoots, and the length and width of the 3rd, 4th, and 5th leaves in each shoot were measured from the day before each application until the first pruning. Relative chlorophyll content was measured one day before each application. The disease index of downy mildew in ‘Kyoho’ grape was surveyed on August 21 and September 3, 2018, and control effect was calculated. Yield per plant, 100-grain weight and soluble solids were measured at harvest.【Results】The average growth of shoot width of ‘Suffolk Red seedless’ and ‘Kyoho’ applied with Lvkangwei and Lvdikang at the same time was 0.123 cm and 0.263 cm, respectively, which were 2.16 and 4.61 times that of the control at the same stage. Shoot length growth in the early stage was greater than the late stage. In the treatment of applying Lvkangwei and Lvdikang at the same time, the shoot length growth was 36.96 cm and 53.82 cm in ‘Kyoho’ and ‘Suffolk Red seedless’, respectively, which was the largest among all treatments, while shoot length increase in the control was only 24.06 cm and 47.91 cm in ‘Kyoho’ and ‘Suffolk Red seedless’, respectively. The effect of microecologics on leaf expansion in the two varieties of grapes was unstable. The chlorophyll content at each stage in the treatments with microecologics application was significantly higher than that in the treatments with no microecologics. Growth increase in both varieties reached the highest value on July 3. The relative chlorophyll content in vines treated with microecologics reached the highest value of 54.2 and 57.8 at time of the sixth application, while water control was only 51.7 and 53.0 in ‘Kyoho’ and ‘Suffolk Red seedless’. ‘Kyoho’ began to develop downy mildew in mid-August. Seven applications of irrigation with Lvdikang and foliage sprays of Lvkangwei during the growing season showed the best control effect on downy mildew in ‘Kyoho’ grape, which was 72.73% and 75.68%. In treatment 4 with the two microecologics applied three times and the fungicides applied three times, the control effect was 54.55% and 49.55%; the control effect with the applications of only Lvdikang for seven times was 58.59% and 57.66%; while the use of the four fungicides was only 60.61% and 38.74%. The yield per plant in ‘Kyoho’ and ‘Suffolk Red seedless’ treated with microecologics was 2.1 and 2.6 times that of the control, and the 100-grain weight was 136.65 g and 67.66 g higher than that of the control, respectively.【Conclusion】Applying microecologics seven times from the leaf-expansion period to the harvesting period to both the above and below ground has a good effect on vine growth and control of downy mildew and can be used as alternatives to chemicals.

Key words: Grape；Microecologics agent; Growth; Grape downy mildew; Yield

霜霉病是葡萄生产过程中最重要的病害之一，在我国最早记载于1899年，各葡萄产区均有发生[1-2]。葡萄霜霉病主要为害叶片，也能通过影响果实成熟而造成严重减产[3]。大量使用杀菌剂仍是目前防治霜霉病的主要措施，CAA类、QoI类等杀菌剂的抗药性已成为葡萄霜霉病防治中的严重问题[2, 4-5]。抗药性的产生一方面需要将杀菌剂轮换使用，一方面激发着新的方向如生物防治的蓬勃发展。

枯草芽孢杆菌（*Bacillus subtilis*）、寡雄腐霉（*Pythium oligorum*）、链孢粘帚霉（*Gliocladium catenulatum*）等已在美国环保局登记用于防治葡萄霜霉病[6]，近年更多新的生防制剂正在研究中[7-10]。笔者选用的绿康威（果树型）和绿地康3号是通过微生态调控对作物发挥促生增产和生物防治作用的微生态制剂，由国家增产菌技术研究推广中心和中国农业大学农用生物制剂中试基地研制，有效成分为枯草芽孢杆菌GLB191、短小芽孢杆菌GLB197、解淀粉芽孢杆菌PG12。为达到经济与环境的双重效益，不少学者对生防制剂和杀菌剂进行了复配或交替使用，对葡萄霜霉病的防治取得了良好的效果[11-12]。有研究显示，一些生防菌在防治葡萄霜霉病的同时，能够促进葡萄生长，增加产量，提高品质[13-14]，壳聚糖等起生防作用的物质则会降低气孔导度而降低光合作用[11]。GLB191和GLB197对葡萄霜霉病有良好防效[15]，而PG12在葡萄上尚未得到应用，GLB191、GLB197、PG12三者混合制剂及其与杀菌

剂交替使用的方法对葡萄促生增产和霜霉病的田间防治效果也未见报道。

为探究微生态制剂在葡萄上应用的最佳方案，在减少化学农药使用的同时，实现葡萄的安全高效生产，笔者在霜霉病发病前用绿康威和绿地康3号对不同栽培管理措施的2个品种的葡萄进行处理。将整个生长季在地上和地下部同时使用微生态制剂的处理与地下部单独处理的效果进行比较；前期使用微生态制剂预防，发病后使用杀菌剂进行治疗，与常规用药相比探究其在减少杀菌剂使用的同时能否达到防病促生效果。以不同处理对葡萄生长量和产量、果实品质的影响和对霜霉病的预防效果为指标，筛选出微生态制剂的最佳使用措施。

1 材料和方法

1.1 材料

供试葡萄品种：‘巨峰’，树龄4 a(年)，露地栽培；‘寒香蜜’，树龄4 a，避雨栽培，由内蒙古自治区包头市果树果品科学技术研究所提供。

供试药剂：绿地康3号(果树型)(下文简称绿地康)，中农绿康(北京)生物技术有限公司，悬浮剂，有效成分为枯草芽孢杆菌GLB191、短小芽孢杆菌GLB197、解淀粉芽孢杆菌PG12，有效活菌数≥10亿•毫升$^{-1}$；绿康威(果树型)(下文简称绿康威)，中农绿康(北京)生物技术有限公司，可湿性粉剂，有效成分为枯草芽孢杆菌GLB191、短小芽孢杆菌GLB197、解淀粉芽孢杆菌PG12，有效活菌数≥200亿•克$^{-1}$；40%百菌清悬浮剂(江阴苏利化学有限公司)；25%甲霜•霜霉威可湿性粉剂(江苏宝灵化工股份有限公司)；70%丙森•多菌灵可湿性粉剂(南通宝叶化工有限公司)。

1.2 方法

1.2.1 试验处理 试验于2018年5—9月在包头市果树果品科学技术研究所开展，第一次用药时‘巨峰’葡萄为展叶期，‘寒香蜜’葡萄为花期前1周，之后每隔10~15 d用药1次，整个生长季施药7次，分别为5月11日、5月23日、6月2日、6月13日、6月23日、7月4日、7月13日。试验设置5个处理，随机区组设计，3个区组，每个处理10株。绿康威与绿地康两者成分相同，菌含量与剂型不同，绿康威粉剂含菌量约为绿地康的20倍，用于地上部处理，绿地康悬浮剂用于地下部处理。处理1为清水对照，叶片正反面均匀喷施清水；处理2为当地常规杀菌剂处理，5月11日—6月2日不用药，6月13日开始4次用药分别为40%百菌清600倍液、25%甲霜灵.霜霉威700倍液、40%百菌清600倍液、70%丙森•多菌灵1 000倍液；处理3使用绿地康500倍液灌根的同时叶面喷施绿康威700倍液处理7次；处理4前4次使用绿地康500倍液灌根的同时叶面喷施绿康威700倍液，后3次与常规用药一致；处理5单独使用绿地康500倍液灌根7次。所有处理的葡萄均在出土后7 d喷施过1次石硫合剂，施肥、灌水、修剪等管理措施均一致。

1.2.2 生长量测量 每次施药前一天统计生长量，直至第一次修剪[13]。每区组随机选3株葡萄，用游标卡尺随机测量上、中、下三个当年生枝的茎粗，卷尺测量茎长[11]。每个枝条选取第3、4、5枚叶片，用直尺测量叶长、叶宽，每次施药前一天用叶绿素测定仪TYS-A(浙江普云农科技股份有限公司)测定相对叶绿素含量[11]。于5月10日、5月22日、6月1日共测量3次，每次测量结果与上一次的差值作为生长量，计算茎粗、茎长、叶长、叶宽的生长量。

1.2.3 各处理对霜霉病的防治效果统计 露地‘巨峰’葡萄八月中旬开始出现霜霉病病状的叶片，于2018年8月21日和9月3日进行病情指数调查和防效计算。每个区组随机选取3株，每株随机调查当年抽生枝蔓上的60枚叶片，采用9级分级法调查发病率和病情指数，计算防效[1, 11]。‘寒香蜜’葡萄无病害发生，不做调查。

9级分级为：0级：无病斑；1级：病斑面积占整个叶面积的5%以下；3级：病斑面积占整个叶面积的6%~25%；5级：病斑面积占整个叶面积的26%~50%；7级：病斑面积占整个叶面积的51%~75%；9级：病斑面积占整个叶面积的76%以上。

$$病情指数=\frac{\sum(各级病叶数\times相应级数)}{调查总叶数\times最高级数}\times100；$$

$$防病效果/\%=\frac{(对照病情指数-处理病情指数)}{对照病情指数}\times100。$$

1.2.4 葡萄产量和品质统计 ‘寒香蜜’葡萄采收时间为8月23日，‘巨峰’葡萄为9月22日，采收时每个区组随机选取3株，用电子天平测量总产量，随机选100粒果实测量百粒质量；每个区组随机选取20个果粒用两层纱布包裹压榨出汁，混匀后用手持折射仪检测可溶性固形物含量[13]。

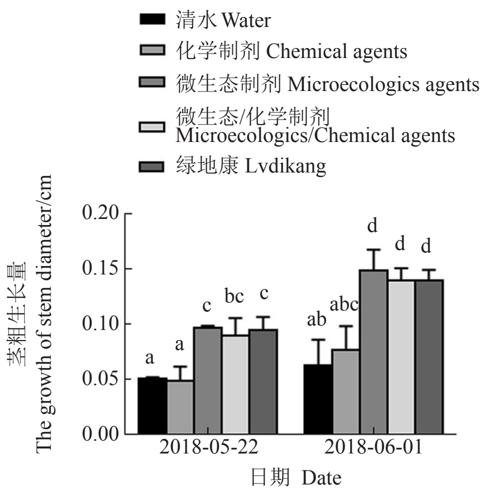
1.3　数据处理

试验数据采用WPS 2019和SPSS Statistics 25.0软件进行统计分析，GraphPad Prism 7软件作图，用Duncan氏新复极差法进行差异显著性检验。

2　结果与分析

2.1　微生态制剂对葡萄的促生效果

由图1~图4可知，同时在地上部和地下部施用微生态制剂的两个处理，‘巨峰’和‘寒香蜜’葡萄茎粗和前期茎长的生长量与单独地下部施用的无显著差别，但都显著大于未使用微生态制剂的，这一趋势在两个品种上显示出了较强的一致性。后期（5月22日—6月1日）茎粗生长量大于前期（5月11日—5月22日），‘巨峰’葡萄和‘寒香蜜’葡萄同时在地上部和地下部施用微生态制剂时茎粗生长量平均值分别为0.123 cm和0.263 cm，为同期清水对照处理的2.16倍和4.61倍。茎长生长则体现为前期大于后期，同时在地上部和地下部施用微生态制剂时两个品种的茎长生长量最大，分别为36.96 cm



图中数据为平均数±标准偏差。不同小写字母表示经Duncan氏新复极差法检验在 $p < 0.05$ 差异显著。下同。

Data in the figure are mean ± SD. Different letters indicate significant difference at 0.05 by Duncan's new multiple range test. The same below.

图1　微生态制剂对‘巨峰’葡萄茎粗生长的影响

Fig. 1　The effect of microecologics agents on the growth of ‘Kyoho’ grape's shoot diameter

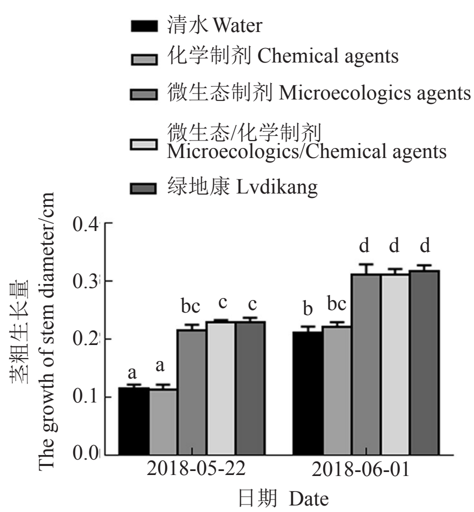


图2　微生态制剂对‘寒香蜜’葡萄茎粗生长的影响

Fig. 2　The effect of microecologics agents on the growth of ‘Suffolk Red seedless’ grape's shoot diameter

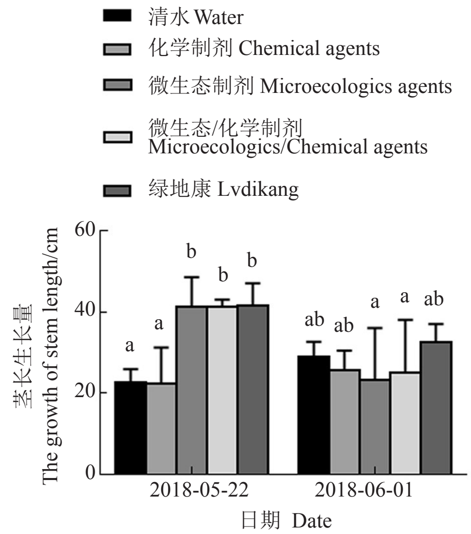


图3　微生态制剂对‘巨峰’葡萄茎长生长的影响

Fig. 3　The effect of microecologics agents on the growth of ‘Kyoho’ grape's shoot length

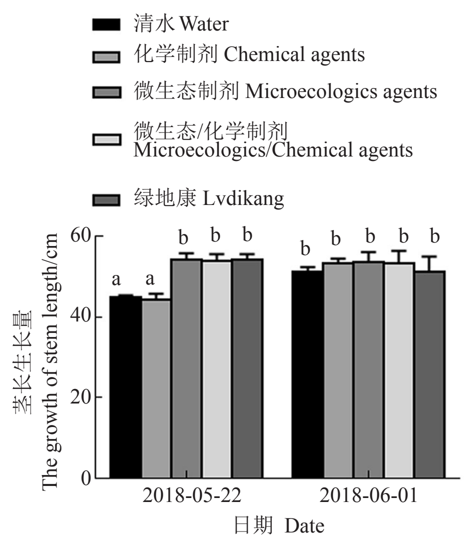
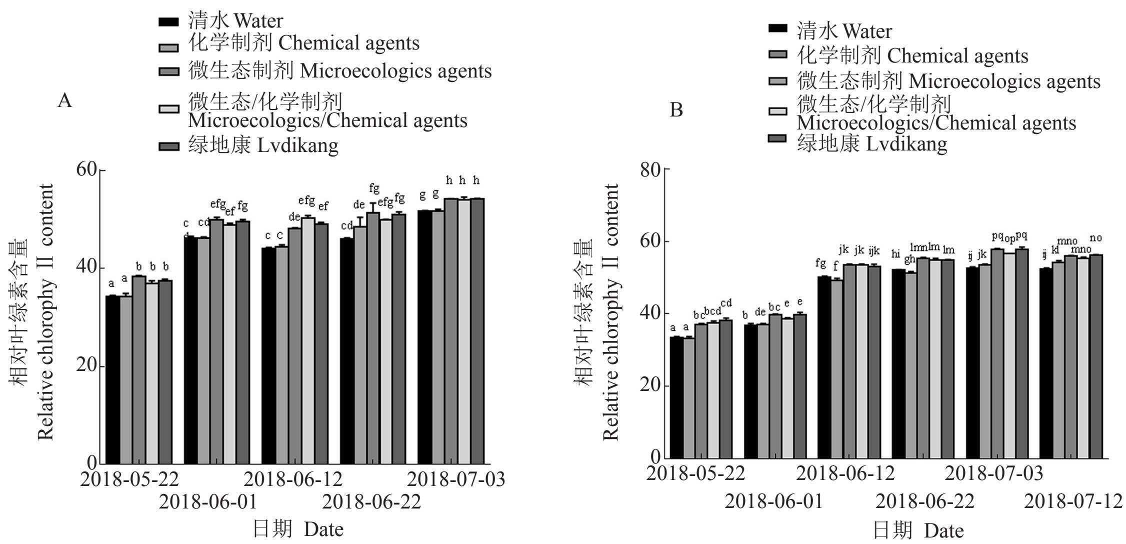


图4　微生态制剂对‘寒香蜜’葡萄茎长生长的影响

Fig. 4　The effect of microecologics agents on the growth of ‘Suffolk Red seedless’ grape's stem length

和53.82 cm，而同期清水对照处理的茎长生长量仅为24.06 cm和47.91 cm。使用微生态制剂对2个品种葡萄的叶长、叶宽生长的影响不稳定，对‘寒香蜜’葡萄叶宽、后期叶长生长有显著促进作用，而对‘巨峰’葡萄无显著影响。

如图5所示，使用微生态制剂的处理各个时期的叶绿素含量显著高于未使用的处理，且两个品种均在7月3日达最高值，地上、地下同时使用微生态制剂处理的‘巨峰’葡萄相对叶绿素含量在第6次处理时达最高值（54.2），‘寒香蜜’达57.8，而清水对照同一时期‘巨峰’葡萄叶绿素相对含量仅为51.7，‘寒香蜜’为53.0。



A. 巨峰；B. 寒香蜜。

A. Kyoho; B. Suffolk Red seedless.

图5 微生态制剂对‘巨峰’和‘寒香蜜’葡萄相对叶绿素含量的影响

Fig. 5 The effect of microecologics agents on relative chlorophyll content of ‘Kyoho’ and ‘Suffolk Red seedless’ grape

2.2 微生态制剂对‘巨峰’葡萄霜霉病的防治效果

如表1所示，‘巨峰’葡萄八月中旬开始出现霜霉病，于8月21日和9月3日进行两次病害调查，调查结果显示出一致的趋势：生长季地上、地下部同时使用微生态制剂处理7次，对‘巨峰’葡萄的霜霉病防效最好，可达72.73%和75.68%；两种微生态制剂处理4次后杀菌剂处理3次，对‘巨峰’葡萄霜霉病的防效为54.55%和49.55%；单独使用绿地康灌根7次，对‘巨峰’葡萄霜霉病的防效为58.59%和57.66%；而使用4次杀菌剂同一时期的防效仅为60.61%和38.74%。第一次调查时单独使用绿地康、杀菌剂和绿地康交替使用与单独使用杀菌剂的处理间无显著差异，第二次调查时单独使用绿地康、杀菌剂和绿地康交替使用的防效均显著高于单独使用杀菌剂处理的防效。

表1 微生态制剂对‘巨峰’葡萄霜霉病的防治效果

Table 1 The control effect of microecologics agents on downy mildew in‘Kyoho’grape

处理 Treatment	病情指数 Disease index (2018-08-21)	相对防效 Control efficacy/% (2018-08-21)	病情指数 Disease index (2018-08-21)	防病效果 Control efficacy/% (2018-09-03)
清水对照 Water control	2.04±0.17 f	-	2.28±0.14 f	-
化学制剂 Chemical agents	0.80±0.04 abc	60.61±0.02 c	1.40±0.05 e	38.74±0.02 a
化学/微生态制剂 Microecologics/Chemical agents	0.93±0.07 cd	54.55±0.03 bc	1.15±0.02 de	49.55±0.01 b
微生态制剂 Microecologics agents	0.56±0.08 ab	72.73±0.04 d	0.56±0.00 a	75.68±0.00 d
绿地康 Lvdikang	0.84±0.02 bc	58.59±0.01 c	0.97±0.03 cd	57.66±0.01 bc

2.3 微生态制剂对葡萄产量和果实品质的影响

巨峰’和‘寒香蜜’葡萄使用过微生态制剂的处理单株产量为清水对照的2.1和2.6倍，使用4次和7次两种微生态制剂、单独使用7次绿地康之

间差别不显著；使用微生态制剂的处理百粒质量高于清水对照136.65 g和67.66 g，但‘巨峰’葡萄中与杀菌剂处理的无显著差别；两个品种的各个处理间可溶性固形物含量无显著差别，如表2、表3所示。

表2　微生态制剂对‘巨峰’葡萄产量和果实品质的影响

Table 2 The effect of microecologics agents on yield and fruit quality in ‘Kyoho’ grape

处理 Treatment	产量 Yield/(kg per plant)	百粒质量 100-grain mass/g	*w*(可溶性固形物) Soluble solids content/%
清水对照 Water control	6.66±0.15 a	845.36±25.20 a	18.7±0.46 a
化学制剂 Chemical agents	8.67±0.62 a	891.84±50.73 ab	18.9±0.24 a
化学/微生态制剂 Microecologics/Chemical agents	14.27±0.65 b	1 035.72±7.92 c	18.3±0.24 a
微生态制剂 Microecologics agents	14.41±1.31 b	982.01±12.50 bc	18.5±0.93 a
绿地康 Lvdikang	12.96±1.21 b	1 072.61±6.98 c	18.7±0.30 a

表3　微生态制剂对‘寒香蜜’葡萄产量和果实品质的影响

Table 3　The effect of microecologics agents yield and fruit quality in ‘Suffolk Red seedless’ grape

处理 Treatment	产量 Yield/(kg per plant)	百粒质量 100-grain mass/g	*w*(可溶性固形物) Soluble solids content/%
清水对照 Water control	0.69±0.06 a	164.03±5.10 a	21.1±0.20 a
化学制剂 Chemical agents	1.17±0.03 b	200.41±8.52 b	20.7±0.33 a
化学/微生态制剂 Microecologics/Chemical agents	1.68±0.10 c	210.30±9.58 bc	20.6±0.09 a
微生态制剂 Microecologics agents	1.81±0.07 c	231.69±4.11 cd	20.6±0.26 a
绿地康 Lvdikang	1.72±0.11 c	240.65±3.35 d	20.6±0.20 a

3　讨　论

葡萄霜霉病在温暖潮湿的气候条件下发生尤为严重，而寒冷地区发病率显著降低，在中国东部和南部地区的发生较为严重[3, 16-17]。‘巨峰’葡萄虽为霜霉病抗性品种[18]，但由于其在露地栽培[19]，霜霉病的发生仍较严重。包头地区降雨集中在7—9月，为‘巨峰’葡萄转色期后，在此期间杀菌剂的使用受到限制，故对该病的控制主要依靠前期的预防。本试验前期使用微生态制剂预防，发病后使用杀菌剂治疗的策略，取得了比常规防治更佳的效果，在减少杀菌剂使用的同时起到促生增产的作用。

霜霉病病原菌为卵菌门葡萄生轴霜霉[*Plasmopara viticola* (Berk. & Curt) Berl. & de Toni][2]，可通过菌丝和吸器大量侵入叶片，因此传统杀菌剂难以控制[20]。包头试验地长期使用广谱性杀菌剂，抗药性的产生是亟待解决的问题，另外杀菌剂的残效期较短，在包头地区的气候条件下难以发挥作用。而葡萄植株表面的微生物可与植株互作，对植株产生有益或致病作用，最终影响葡萄的产量和品质[21]。GLB191通过丰原素和表面活性素的产生与病原体直接作用，通过诱导防御基因表达和胼胝质产生诱导植物抗病性发挥作用，从而防控葡萄霜霉病的发生[22]。PG12主要通过伊枯草素和丰原素发挥广谱的抗菌活性[23]。本试验中芽孢杆菌在葡萄根部和叶部的定殖使得植株表面的微生物群落结构改善，其次生代谢物促进植株生长，增强树势，使植株对霜霉病抗性增强，并提高产量，且整个生长季持续用药使得该效应持续时间较长。

4　结　论

在霜霉病发病较轻的地区，在地上和地下部同时使用微生态制剂对葡萄霜霉病有良好防效，对葡萄还有促生增产作用，具有作为杀菌剂的替代在田间使用的潜力。

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