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环渤海湾苹果产区老果园与连作果园土壤线虫群落特征

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摘要:应用类群多样性、类群丰富度、个体密度和功能类群指数等群落参数,于2009年5—10月分3次对环渤海湾苹果栽植区老果园不同位置和连作果园取样研究了土壤线虫群落特征。结果表明:苹果连作对果园土壤生物环境具有明显的恶化作用,连作果园土壤环境条件恶劣;对功能类群指数的统计表明,苹果连作明显改变了果园土壤中植物寄生性线虫 *r*-选择和 *k*-选择的比例,与自由生活线虫比较,连作对植物寄生线虫影响更明显;连作提高了果园的土壤线虫 *PPI/MI*,说明连作对果园土壤健康的扰动最大;土壤不同食性线虫数量统计结果显示,环渤海湾苹果栽植区植食性线虫数量未达线虫伤害阈值,线虫不是引起环渤海湾地区苹果连作障碍的主要原因。

关键词:土壤线虫;连作果园;环渤海湾

Characterization of nematode communities in the soil of long-standing versus replanted apple orchards surrounding Bohai Gulf

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Abstract: Apple replant disease (ARD) is a common problem affecting the reuse of lands just removed from apple production. ARD problem is caused by soil-borne organisms, which stunts the growth of apple trees, suppresses the yield and quality of apple fruits, and enhances susceptibility to diseases and pests. Apple replanting impacts the pH value, nutrition level, enzymatic activity, concentration of toxic substances (e. g. phenolic compounds), and microorganism community (e. g. pathogenic fungi) in the soil, deteriorating physical and chemical properties of soil. Healthy soil is essential for producing healthy foods and protecting human health. Soil nematodes (*Phylum nematoda*) are important components in the soil ecosystem, play significant roles during the processes of soil formation and maturation, and are major parts in the soil food chain. Soil nematodes are sensitive to physical and chemical changes in the soil, and are often used as the biological indicator measuring soil quality. In this study, soil nematodes were analyzed on apple orchards surrounding Bohai Gulf from May to October in 2009. The structure and dynamic change of nematode communities were assessed by using generic diversity, generic richness, nematode density, and a functional group index. In comparison, the generic diversity, generic richness and uniformity of soil nematodes in the replanted apple orchards were significantly lower than those in the long-standing apple orchards. Data of generic diversity also suggested that apple replanting impacts the biological properties of the soil, leading to a less favorable soil environment for apple growth. The maturity index (*MI*) of free-living nematodes was similar between long-standing and replanted apple orchards, but the maturity index of plant parasitic nematodes, or the plant parasite index (*PPI*), was significantly higher in the replanted apple orchards than those in the long-standing apple orchards. According to the functional group index, apple replanting significantly changed the

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ratio of *r*- and *k*- selection of plant parasitic nematodes. When compared with free-living nematodes, plant parasitic nematodes were more adaptable to replanted apple orchards, increasing the *PPI/MI* ratio. Analysis of nematodes with different feeding habits revealed that the overall numbers (36.8 nematodes per 100g dry soil) of herbivorous nematodes in replanted orchards around Bohai Gulf was greatly below the density threshold (114 nematodes per 100g dry soil) that can damage apple trees. Therefore, it appears that nematodes are not the major causal agent for apple replant disease in the Bohai Gulf area. However, it is still possible that soil nematodes might boost the ARD problem in some replanted apple orchards.

Key Words: apple replant disease; Bohai gulf; *Phylum nematoda*

环渤海湾地区是我国苹果主要栽植区,但大片果园开始老化,需进行果园更新,由于土地资源有限,苹果连作普遍存在,由此引起的苹果连作障碍成为果园更新的主要限制因素。

土壤健康是农产品安全和人类健康的基础,土壤生物作为土壤变化的早期预警生态指示,能敏感的反映土壤健康变化过程[1]。线虫是土壤生态系统的重要组成部分,在土壤形成与熟化过程中起着重要作用[2-5]。土壤线虫在土壤食物网中具有重要作用,并且对土壤质量变化反应敏感,常用来作为敏感性指示生物[6-7]。

有研究认为植食性线虫是引起苹果连作障碍的原因之一[8-9],也有研究结果显示根结线虫不是引起苹果连作障碍的原因[10]。

因此,在环渤海湾苹果栽培区多点定位取样,研究连作果园土壤线虫群落特征,以期为生产上采取适当措施防止苹果连作障碍提供依据。

1 材料与方法

1.1 试验区概况

渤海湾位于渤海西部,是三面环陆的半封闭性海湾,位于中纬度季风区,气候有显著“大陆性”特征:一是季风显著;二是冬寒夏热,四季分明,春秋短促,气温年变差大;三是雨季很短,集中在夏季,7、8 两月降水量占全年的 64%—68%,春季少雨,降水量的年际变化也很大。

1.2 土样采集与线虫测定

分别于 2009 年 4 月 28 日—5 月 1 日(春季)、7 月 26 日—28 日(夏季)和 10 月 9 日—11 日初(秋季)共 3 次在环渤海湾苹果栽植区山东的昌邑、栖霞、蓬莱,辽宁绥中、瓦房店、锦州,河北的昌黎、扶宁 8 个地点进行土壤取样,每个点分别设老果园行内、行间及连作果园行内 3 个不同取样点,每个不同取样点设 3 次重复,取样深度为 20cm,采集用于土壤线虫的样品共 216 份,采用淘洗-过筛-蔗糖离心法实验室分离提取土壤线虫[11],线虫标本依据尹文英等《中国土壤动物检索图鉴》[12]鉴定,并根据土壤湿度,将土壤线虫个体数量换算成 100g 干土中含有的线虫数目。

1.3 数据统计与处理

采用 *MI*(maturity index)指数、*PPI*(plant-parasite index)指数和 $\sum MI$ 指数表达各生境土壤线虫群落功能结构特征[13-14]。

上述 3 个指数形成关系公式相同,为[15-16]:

$$\sum MI(PPI) = \sum cp_i \times p_i$$

式中,cp_i为土壤自由生活(植物寄生性)线虫第 i 类群 colonizer-persister(*cp*)值;n 为土壤自由生活(植物寄生性)线虫类群数;p_i为土壤线虫群落自由生活(植物寄生性)线虫第 i 类群的个体数占群落总个体数的比例。

运用 Shannon-Wiener 指数(H')、Simpson 指数、Pielou 均匀度指数(J)和 Margalef 丰富度指数(SR)相结合研究土壤线虫群落的多样性[3,17-19]。

$$H' = -\sum p_i \times \ln p_i$$

$$D = 1 - \sum P_i^{\,2}$$

式中,p_i为土壤线虫群落第 i 类群个体数占总个体数比例;S 为土壤线虫类群所有线虫类群数。

$$J = H'/\ln S$$

$$SR = (S - 1)/\ln N$$

式中,N 为土壤线虫群落全部类群的个体总数。

根据线虫头部形态学特征和取食生境将线虫分为 4 个功能(营养)类群:食细菌类群、食真菌类群、植食性线虫、捕食类群/杂食类群[18,20]。

数据统计采用 DPS 统计分析软件。

2 结果与分析

本研究共捕获土壤线虫 59180 条,个体平均密度 243.5/100g 干土,其中,春季、夏季和秋季取样线虫数目分别为 13 899、25 125 和 13 356 条,分别占取样总量的 26.5%、48%、25.5%,在山东、辽宁和河北线虫取样数目分别为 10 538、10 915 和 30 941 条,分别占取样总量的 20.1%、20.8%、59.1%。

2.1 环渤海湾苹果产区老果园与连作果园土壤线虫群落结构

环渤海湾各地区土壤线虫群落结构如表 1 所示。线虫个体密度差异显著。除样地 LN 行间和 LN 连作的丰富度 SR 指数较大外,不同取样地点老果园和连作果园相比,土壤线虫类群数、多样性 H'指数、D'指数,均匀度 J 指数和多样性 SR 指数均表现出连作果园低于老果园,其中,土壤线虫类群数、个体密度、多样性 H'指数、D'指数均有显著不同,J 指数和丰富度 SR 指数差异不大。总体上看,除个别取样地点,以上所有群落结构指标老果园行间最高,连作果园差异较低。

表 1 不同地区老果园与连作果园土壤线虫群落结构

Table 1 Structure of soil nematode community in replant orchard and old orchard in different regions

取样位置 Sampling position	类群数 Genus number	个体密度 Density	多样性 Diversity H'	多样性 Diversity D'	均匀度 Evenness J	丰富度 Richness SR
SD 行间	11.56 ±1.06a	174.4 ±11.5e	1.43 ±0.02ab	0.67 ±0.07ab	0.63 ±0.05b	1.75 ±0.15a
SD 行内	8.67 ±0.44cd	243.2 ±3.5d	1.1 ±0.01cd	0.45 ±0.03d	0.45b	1.34 ±0.05b
SD 连作	6.22 ±0.22ef	284.7 ±11.7c	1.01 ±0.06d	0.45 ±0.03d	0.49 ±0.01b	1.25 ±0.08b
LN 行间	11.17 ±0.17ab	359.4 ±9.6b	1.59 ±0.1a	0.75 ±0.05a	1.23 ±0.46a	1.89 ±0.03a
LN 行内	9.56 ±0.33bc	311.3 ±1.6c	1.18 ±0.02cd	0.46d	0.60 ±0.01b	1.35 ±0.06b
LN 连作	9.94 ±0.83abc	134.7 ±0.13f	1.43 ±0.06ab	0.59bc	0.59 ±0.01b	1.89 ±0.16a
HB 行间	7.08 ±0.08de	89.7 ±1.6g	1.26 ±0.09bc	0.5cd	0.59 ±0.17b	1.34 ±0.16b
HB 行内	9.33 ±0.33c	389.0 ±18.9a	1.32 ±0.11bc	0.56 ±0.02ed	0.55 ±0.01b	1.57 ±0.07ab
HB 连作	5.25 ±0.83f	351.117.6b	0.57e	0.26 ±0.01e	0.35b	0.84 ±0.01c

注:不同字母含义 SD:山东,LN:辽宁,HB:河北; 同一列中不同小写字母表示达到 5% 水平差异显著(邓肯法)

表 2 不同季节老果园与连作果园土壤线虫群落结构

Table 2 Structure of soil nematode community in replant orchard and old orchard in spring, summer, autumn

季节 Season	类群数 Genus number	个体密度 Density	多样性 Diversity H'	多样性 Diversity D'	均匀度 Evenness J	丰富度 Richness SR
春季 Spring	8.79 ±0.28b	325.92 ±80.8b	1.57 ±0.02a	0.54 ±0.01a	0.59b	1.16 ±0.06a
夏季 Summer	8.34 ±0.04b	288.22 ±7.99b	1.33 ±0.05b	0.54b	0.56 ±0.02c	1.13 ±0.06b
秋季 Autumn	16.06 ±0.85a	521.38 ±31.06a	1.20c	0.77 ±0.07b	1.02 ±0.32a	1.12 ±0.07b

同一列中不同小写字母表示达到 5% 水平差异显著(邓肯法)

综合所有样地,不同季节对土壤线虫群落结构的影响如表 2 所示,土壤线虫类群数、个体密度、多样性 D'指数、均与度 J 指数均呈现秋季最高,而多样性 H'指数和丰富度 SR 指数随季节变化呈下降趋势,春季最高,秋季最低,春秋两季呈显著性差异。

2.2 环渤海湾苹果产区老果园与连作果园土壤线虫群落功能类型特征

采用∑*MI* 指数、*PPI* 指数和 *PPI/MI* 值研究不同地区再植果园和老果园土壤线虫功能结构的差异如表 3 所示,山东、辽宁、河北 3 个地区连作果园∑*MI* 指数分别为 1.13、1.21 和 3.04,*PPI* 指数分别为 0.47、0.57 和 0,与对应的老果园行内和行间相比具有较高的∑*MI* 指数和较低的 *PPI* 指数,除山东∑*MI* 指数未到达显著性差异外,辽宁和河北两地连作果园和老果园相比,∑*MI* 指数有显著差异;除河北省外,其他两个地区连作果园的 *PPI/MI* 指数低于老果园行间,而高于老果园行内的 *PPI/MI* 指数。

表 3 不同地区老果园与连作果园土壤线虫功能类群特征

Table 3 Characteristic of functional group of soil nematode in replant orchard and old orchard in different regions

取样位置 Sampling position	自由生活线虫成熟度指数 ∑*MI*(maturity index)	植物寄生线虫成熟度指数 *PPI*(plant-parasite index)	*PPI/MI*
SD 行间	1.07 ±0.02bc	0.75 ±0.04b	1.0 ±0.06b
SD 行内	1.24bc	0.33 ±0.04cd	0.29 ±0.01cde
SD 连作	1.13 ±0.03bc	0.47 ±0.05cd	0.33 ±0.03cd
LN 行间	1.48 ±0.63b	1.02 ±0.19a	2.07 ±0.21a
LN 行内	0.63 ±0.01c	0.35 ±0.04cd	0.19 ±0.08de
LN 连作	1.21 ±0.01bc	0.57 ±0.02bc	0.49 ±0.04c
HB 行间	1.4 ±0.01b	0.31d	0.21 ±0.08cde
HB 行内	1.70 ±0.02b	0.36 ±0.06cd	0.15 ±0.03de
HB 连作	3.04 ±0.06a	0	0

由表 4 可以看出,∑*MI* 指数、*PPI* 指数和 *PPI/MI* 值不同季节存在差异,春季、夏季、秋季呈升高趋势,春季和秋季两季节呈显著性差异。

表 4 不同季节老果园与连作果园土壤线虫功能类群特征

Table 4 Characteristic of functional group of soil nematode in replant orchard and old orchard in spring, summer, autumn

季节 Season	成熟度指数 ∑*MI*(maturity index)	植物寄生线虫成熟度指数 *PPI*(plant-parasite index)	*PPI/MI*
春季 Spring	0.07 ±0.07b	0.37 ±0.01b	0.37 ±0.01b
夏季 Summer	1.39 ±0.03ab	0.61 ±0.09ab	0.59 ±0.1b
秋季 Autumn	3.01 ±0.17a	0.75 ±0.04a	1.65 ±0.25a

2.3 环渤海湾苹果产区老果园与连作果园土壤中线虫食性类群

由表 5 可以看出,不同地区线虫食性类群数量明显不同,其中河北果园内食细菌性线虫数量最高,杂食-捕食性线虫数量最低,而山东果园食细菌性线虫数量最低,杂食-捕食性线虫数量最高;各地区果园土壤中食真菌性线虫数量变化无明显差异;在所有土样中,山东老果园行内植食性线虫数量最高,达 58.1 条/100g 干土,连作果园土壤中植食性线虫数量以辽宁最高,为 36.8 条/100g 干土。

表 5 不同地区老果园与连作果园土壤线虫各食性类群密度/(条/100g 干土)

Table 5 Number of different feeding habits of soil nematode in replant orchard and old orchard in different regions

取样位置 Sampling position	食细菌性线虫 Bacterivores	食真菌性线虫 Fungivores	植食性线虫 Plant-parasites	杂食-捕食性线虫 Predators/Omnivores
SD 行间	74	9.4	46	35.2
SD 行内	58	11.8	58.1	19.7
SD 连作	42.4	7.8	14.2	13.9
LN 行间	20.6	5.5	15.9	14.2
LN 行内	118.9	8.4	8.6	11.7
LN 连作	133.5	10.5	36.8	19.5
HB 行间	453.1	18.9	44.4	3.3
HB 行内	957	7.4	8.5	4.1
HB 连作	219	0.9	0.4	1.9

由表 6 可以看出,山东、辽宁、河北 3 个地区不同取样部位土壤各食性线虫数量均以夏季最高,夏季是线虫活动的高发季节。

表 6　不同季节老果园与连作果园土壤线虫各食性类群数密度/(条/100g 干土)

Table 6　Number of different feeding habits of soil nematode in replant orchard and old orchard in spring, summer, autumn

季节 Season	食细菌性线虫 Bacterivores	食真菌性线虫 Fungivores	植食性线虫 Plant-parasites	杂食-捕食性线虫 Predators/Omnivores
春季 Spring	151.9	7.9	26.3	13.8
夏季 Summer	271.1	13.5	34.2	16
秋季 Autumn	154.6	5.2	20.4	15.3

3　讨论

土壤线虫是土壤中最丰富的土壤动物,也是最重要的次级消费者,已被作为土壤生物多样性和功能的指示生物[21-24]。

生物多样性是群落生物组成结构重要指标,反映群落内物种多少和生态系统食物网复杂程度,从而反映生境间相似性及差异性。综合本研究所有取样地区结果显示,老果园行间土壤线虫拥有较高的多样性,均匀度和丰富度,老果园行内和连作果园土壤线虫多样性、均匀度和丰富度显著降低,连作果园最低,说明连作果园土壤环境条件恶劣,苹果连作对果园土壤生物环境具有恶化作用。

MI 指数和 *PPI* 指数最早由 Bongers 提出[15],用以反映土壤线虫群落功能结构特征。*MI* 指数和 *PPI* 指数分别指示土壤自由生活线虫和植物寄生线虫 *r*-选择和 *k*-选择的比例,显示线虫的生活周期、繁殖能力和抗干扰能力的强弱[25]。后来,Yeates 等又提出了 $\sum MI$ 指数[16]。本研究发现,环渤海湾各地区不同取样部位均表现老果园行间 *PPI* 指数最高,连作果园次之,老果园行内最低,而 $\sum MI$ 指数差异不大,表明栽植果树和苹果连作明显改变了果园土壤中植物寄生性线虫 *r*-选择和 *k*-选择的比例,与自由生活线虫相比,植物寄生线虫对栽植果树更敏感。

人为扰动会使土壤线虫群落 *PPI/MI* 值升高,未受扰动的自然环境线虫 *PPI/MI* 值将低于扰动环境[26],在一定条件下 *PPI/MI* 值反映土壤生态系统对外界干扰恢复程度可能更敏感。本研究发现环渤海湾地区连作果园土壤线虫 *PPI/MI* 值低于同地区老果园行间且高于老果园行内,与 Bongers 等的上述推断相似。老果园行内由于施肥等管理外界人为活动扰动多,表现出最低的 *PPI/MI* 值;老果园行间由于果树的长期生长人为对老果园土壤的扰动最少,*PPI/MI* 值最高;而连作果园内,虽然初期存在刨除老树、整地、施肥、栽植幼树,但栽植后期人为对土壤线虫扰动减少,所以 *PPI/MI* 值位于两者之间。比较不同季节对环渤海湾苹果栽植区果园土壤线虫 *PPI/MI* 值表明,果树生长降低了土壤线虫 *PPI/MI* 值,随着果树生长时间的延长,*PPI/MI* 值呈下降趋势。

国内外众多研究对土壤线虫与苹果连作障碍关系的结论不一致,有研究结果表明植食性线虫是苹果再植病的原因之一[27-29]。郝玉金等研究发现南京毛刺线虫(*Trichodorus nanjing ensis* Liu & Chen)是导致苹果树势衰退、再植病和短粗艮病的主要病原[30]。也有结果认为根结线虫不是引起苹果再植病的原因[31-33]。阮维斌等 1999 年对四川 27 县果园线虫发生情况调查认为四川省苹果连作障碍与线虫有关,且线虫是主要致病因子[34]。土壤线虫对苹果连作障碍的影响可能随地区不同而不同。植食性线虫密度作为影响苹果连作障碍的指标已被广泛应用[28-29,31,33]。本研究对环渤海湾地区苹果栽植区果园土壤中各食性线虫数量统计结果表明,植食性线虫数量远没有达到植物伤害线虫阈值(114 条/100g 干土)[27],可以推断线虫不是阻碍该地区苹果园更新的主要因子。

土壤线虫处于土壤环境中,其消长受气候[35]、土壤动物、微生物[36]以及其他类群线虫[37]的影响,此外土壤有机质[38]、孔隙度[39]、温湿度、通气也是影响线虫分布的因素[40]。

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