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A preliminary study on the root-knot nematode resistance of the cherry plum cultivar Mirabolano 29C

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Abstract: Grafting is the most important means of breeding peach, plum, apricot, and other fruit trees, and the selection of the rootstock is crucial to the quality of the grafting and the yield of the products. The traditional commonly used peach rootstock is susceptible to root-knot nematode infections, resulting in a decreased yield, while a variety of cherry plum rootstocks, Mirabolano 29C, is resistant to root-knot nematode. In this study, root-knot nematode infection experiments on seedlings of traditional peach rootstocks and Mirabolano 29C confirmed that Mirabolano 29C was indeed more resistant to root-knot nematodes. At the same time, we compared the roots of the root-knot nematode uninfected and infected Mirabolano 29C by transcriptome sequencing and found 3 176 differentially expressed genes. A further functional enrichment analysis of these genes found that the secondary metabolites, phenylpropane and flavonoids, may be responsible for the high resistance of Mirabolano 29C to root-knot nematodes. These results can provide a reference value for the disease resistance breeding of rootstocks.

Keywords: disease resistance; grafting; phenylpropane pathway; rootstocks; transcriptome sequencing

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Grafting is an ancient method that has commonly been applied in agriculture for thousands of years and it has many advantages (Notaguchi et al. 2020; Guo et al. 2017; Thomas & Frank 2019). For example, plants often show higher abiotic and biotic resistance after grafting compared with their parents and they can even live in a new environment (King et al. 2008; Melnyk & Meyerowitz 2015; Guo et al. 2017). In addition, grafting is also used to improve the yield in agriculture and to change the branch structure (Thomas & Frank 2019). Nowadays, it is an effective way to propagate many fruit trees with an economic and nutrient value like the peach (*Prunus persica* L.), plum (*Prunus salicina* Lindl.), and apricot (*Prunus armeniaca*) by grafting (Notaguchi et al. 2020).

The rootstock is an important part of the grafting of fruit trees. Thus, the types of rootstocks, their physiological activities, and their ability to resist stress, are the factors that we need to consider when choosing rootstocks. Currently, pests, especially root-knot nematodes, are thought to be one of the main factors affecting the agricultural yield and quality (Dodds & Rathjen 2010; Xu et al. 2019; Forghani & Hajihassani 2020). Reducing the losses from root-knot nematodes through some pesticides is effective, but this practice is not supported as it will undoubtedly put huge pressure on the environment (Xu et al. 2017). Therefore, it is an effective way to breed rootstocks that are more resistant to pests and diseases.

Peaches, plums, apricots and other stone fruit agricultural products are widely loved by people for their rich nutritional value, sweet and refreshing taste and great economic value (Faust et al. 1998; Byrne et al. 2012; Birwal et al. 2017). Grafting is the main way of propagation, so the selection of rootstocks is very important. The rootstock of peaches, as the common rootstock for grafting, are susceptible to root-knot nematode infections (Lu et al. 1998). When root-knot nematodes infect a plant, many root galls form on the main root and lateral roots of the plant, which seriously affect the transport of nutrients and cause poor plant growth and development, causing great losses to production (Opperman et al. 1994). Mirabolano 29C (*Prunus cerasifara*) is a rootstock native to the United States of America and is used for drupe fruit trees, such as plums and apricots, in Europe and the United States of America. It has strong resistance to root-knot nematodes. However, there is no report on the molecular level of its resistance to root-knot nematodes. In this study, we compared the resistance of the peach rootstock and Mirabolano 29C to root-knot nematodes and explored the potential reasons that Mirabolano 29C shows resistance to root-knot nematodes by transcriptome sequencing. The obtained results provide a theoretical basis for research on rootstock resistance to nematodes and diseases.

MATERIAL AND METHODS

Preparation and treatment of plant materials. Seeds of cherry plums (Mirabolano 29C) (*Prunus cerasifara*) and peaches (*Amygdalus persica* (L.) Batsch) were sown to grow normally in soil free of root-knot nematodes (*Meloidogyne incognita*, (GH-1)). One year later, 30 well-growing cherry plum and peach plants were selected for the subsequent experiments. Half of the seedlings of each material were transplanted into a root-knot nematode-rich (second instar larvae, about 500 nematodes/100 g) soil. A mixture of lateral and fibrous roots of Mirabolano 29C grown normally and treated with nematodes for three days was taken for transcriptome sequencing. Three biological replicates of each group were used. In addition, fifteen days later, the nematode infestation of the roots of both materials was compared.

Total RNA extraction, RNA-Seq library preparation and sequencing. TRIzol (Rio et al. 2010; Zhang et al. 2010) was used for RNA extraction. The mRNA from three organs (three replicates) was purified from the total RNA using oligo (dT) magnetic beads for sequencing (Young et al. 2010; Patterson et al. 2019; Rao et al. 2020). A DNA probe was digested with DNase I after hybridisation with rRNA to obtain purified RNA. A fragmentation buffer was then used to turn the RNA into small fragments. Random N6 primers were used to synthesise the first-strand cDNA, which was then followed by a second-strand cDNA synthesis. The ends of the double cDNA were repaired; the 5' ends were phosphorylated, and the 3' ends were A-tailed to form cohesive ends (Beijing Genomics Institute, Shenzhen, China). Then, the cDNA was ligated to the sequencing adapters. The ligation products were amplified using polymerase chain reaction (PCR) to build a cDNA library which was sequenced on the DNBSEQ-T7 platform (Beijing Genomics Institute, Shenzhen, China). The raw RNA sequence data were submitted to the National Center for Biotechnology Information (NCBI) Sequence Read Archive with accession number PRJNA809767.

Transcript annotation and novel transcript prediction. To obtain clean reads after the RNA-seq, raw reads containing sequencing adapters with more

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than a 5% unknown base or more than a 20% low-quality base were removed using SOAPnuke software (Ver. 2.1.0) (Chen et al. 2018). STAR (Ver. 2.7.6a) was used to map the clean reads to the reference genome of Mirabolano 29C (https://www.rosaceae.org/Analysis/9450778) from the GDR (Genome Database for Rosaceae). To predict the potential novel transcripts, StringTie (Ver. 1.2.4) was used to align each sample to the genome, redundantly removing and filtering out transcripts that overlap with known transcripts to obtain a new set of transcripts, and the Coding Potential Calculator (CPC) was used to predict the coding capacity of the new transcripts (Pertea et al. 2016).

Differential expression analysis and functional enrichment. RSEM (Ver. 1.2.30) (Li & Dewey 2011) was used to estimate the expression values for each sample, and the read counts were normalised using the fragment per kilo-base of transcript per million fragments mapped (FPKM). The R package DESeq2 (Ver. 1.28.1) was used to perform the differential expression analysis on the read-count data (Love et al. 2014). Genes with $|\log_2$ (fold change)$|$ > 1 and Benjamini-Hochberg (B & H) corrected false discovery rate (FDR) < 0.05 were considered differentially expressed genes (DEGs). For a visual comparison of each sample, the R package ggplot2 (Ver. 3.3.0) was used to construct a volcanic map (Wickham 2011). The DEGs were subjected to a gene ontology (GO) (Ashburner et al. 2000) functional enrichment analysis and a Kyoto Encyclopedia of Genes and Genomes (KEGG) (Kanehisa et al. 2004) analysis using the R package clusterProfiler for functional annotation (Ver. 3.16.1) (Yu et al. 2012). The GO terms and KEGG pathways with $P < 0.05$ were defined as significantly enriched in DEGs.

RESULTS

Comparison of two rootstocks. As shown in Figure 1, when treated with nematodes, the root system of the peach was infected and there was obvious swelling. However, the roots of Mirabolano 29C showed no difference between the group grown under non-infected conditions and that treated with nematodes. In this part, we can suggest that the roots of Mirabolano 29C have stronger resistance to nematodes than the peach. Thus, to discover the reason, we performed a transcriptome analysis on two groups (CK and treated with nematodes) of the Mirabolano 29C roots.

Sequencing and mapping. Six different raw data of the Mirabolano 29C samples (three CK: root without nematode treatment; three CT: root with nematode treatment) were generated by next-generation sequencing. After filtering the raw data, clean reads ranging from 47 650 524 to 98 110 150 were generated (Table 1). The RNA sequence mapping revealed that 92.83–94.90% of the clean reads could be mapped onto the reference genome, with most of them being uniquely mapped (Table 2). In addition, 14 468 *novo* transcripts were predicted (Table S1 in the Electronic Supplementary Material (ESM)).

Differentially expressed genes and function enrichment. To reveal the differences between CK and

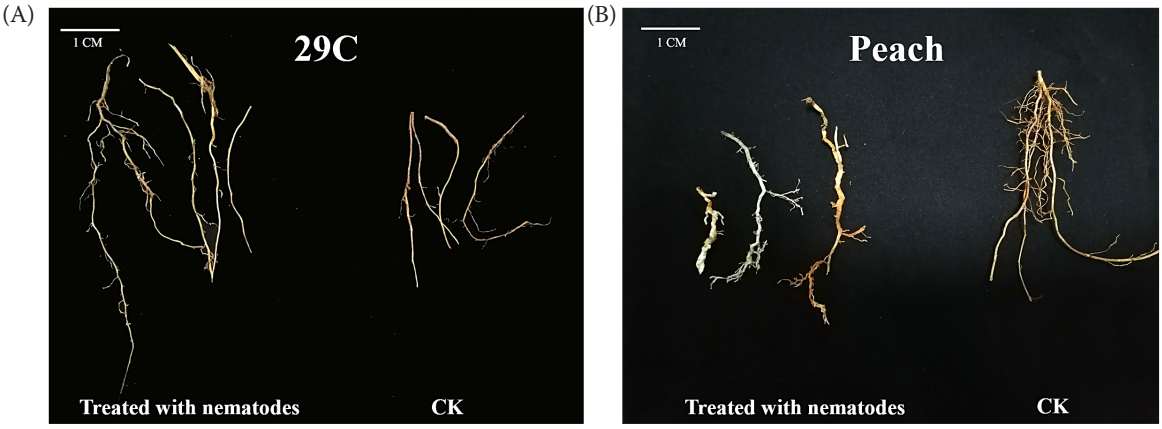

Figure 1. Mirabolano 29C roots treated with root-knot nematodes for 15 days (A left), Mirabolano 29C roots under normal growth conditions (CK) (A right), peach roots treated with root-knot nematodes for 15 days (B left), peach roots under normal growth conditions (CK) (B right)

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Table 1. Summary of the RNA-Seq data and quality of sequencing

Sample	Raw data	Clean data	Clean data ratio	Clean data Q20	Clean data Q30	GC content
			(%)			
CK-1	63 090 204	62 054 888	98.36	96.31	89.44	46.40
CK-2	99 873 796	98 110 150	98.23	96.03	88.76	47.71
CK-3	91 699 272	90 164 324	98.33	96.10	88.98	46.73
CT1-1	48 455 606	47 650 524	98.34	96.03	88.73	46.09
CT1-2	79 436 312	78 433 388	98.74	95.59	87.47	46.33
CT1-3	91 964 394	90 711 424	98.64	95.76	87.87	46.02

Q20 – the percentage of bases with a Phred value > 20; Q30 – the percentage of bases with a Phred value > 30; GC content –the percentage of nitrogenous bases in a DNA or RNA molecule that are either guanine (G) or cytosine (C)

Table 2. Six transcriptome samples mapped to the Genome

Sample	Total clean reads	Mapped reads	Uniquely mapped reads	Unmapped reads
CK-1	62 054 888	58 889 540 (94.90%)	52 164 068 (84.06%)	3 165 348 (5.10%)
CK-2	98 110 150	92 956 474 (94.75%)	68 431 316 (69.75%)	5 153 676 (5.25%)
CK-3	90 164 324	85 240 046 (94.54%)	71 275 066 (79.05%)	4 924 278 (5.46%)
CT1-1	47 650 524	44 908 100 (94.24%)	40 753 590 (85.53%)	2 742 424 (5.76%)
CT1-2	78 433 388	72 809 996 (92.83%)	65 783 294 (83.87%)	5 623 392 (7.17%)
CT1-3	90 711 424	85 337 328 (94.08%)	77 339 892 (85.26%)	5 374 096 (5.92%)

CT, we performed a comparative analysis of the six samples. The DEGs were determined at a threshold of $|\log_2(\text{fold change})| > 1$ and FDR < 0.05. There were 3 176 genes that differentially expressed when comparing the CT to CK group (Table S2 in the ESM), with 987 up-regulated and 2 189 down-regulated genes (Figure 2).

The biological processes, cell components, and molecular functions connected with these DEGs are intuitively reflected by the enrichment of the GO terms of the different genes. The DEGs found in each group were subjected to a GO analysis, and the enriched GO terms were recorded (Table S3 in ESM). For example, DEGs were significantly enriched into cellulose biosynthetic process, cellular glucan metabolic process, xyloglucan metabolic process, and cell wall biogenesis. The top enriched 20 terms are shown in Figure 3.

The DEGs were also processed through a KEGG pathway analysis, which is a different way of categorising the gene activity that focuses on biochemical pathways. Figure 4 depicts the top 20 KEGG pathway enrichment analyses while there were nine pathways that the DEGs insignificantly enriched, which were phenylpropanoid biosynthesis, diterpenoid biosynthesis, carotenoid biosynthesis, galactose metabolism, glutathione metabolism, ABC transporters, pentose and glucuronate interconversions, flavonoid biosynthesis, and zeatin biosynthesis (Table S4 in the ESM).

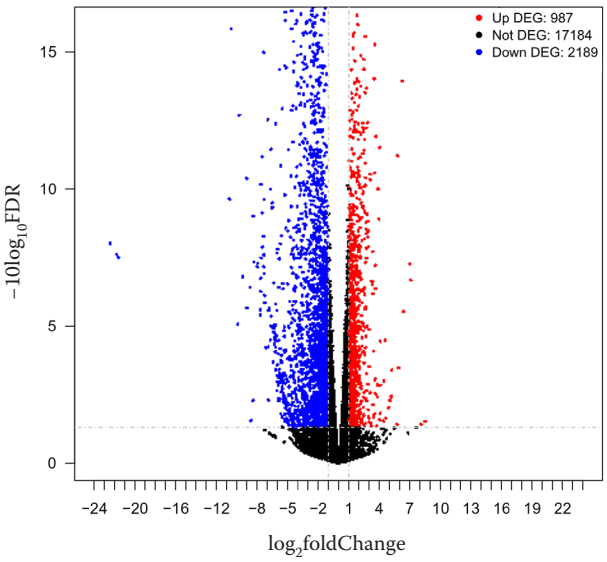

Figure 2. Volcano plots of the differentially expressed genes (DEG) between the roots with the nematode treatment (CT) and roots without the nematode treatment (CK) group
FDR – false discovery rate; the blue and red dots represent genes that are significantly down-regulated and up-regulated in the CT relative to the CK group, respectively

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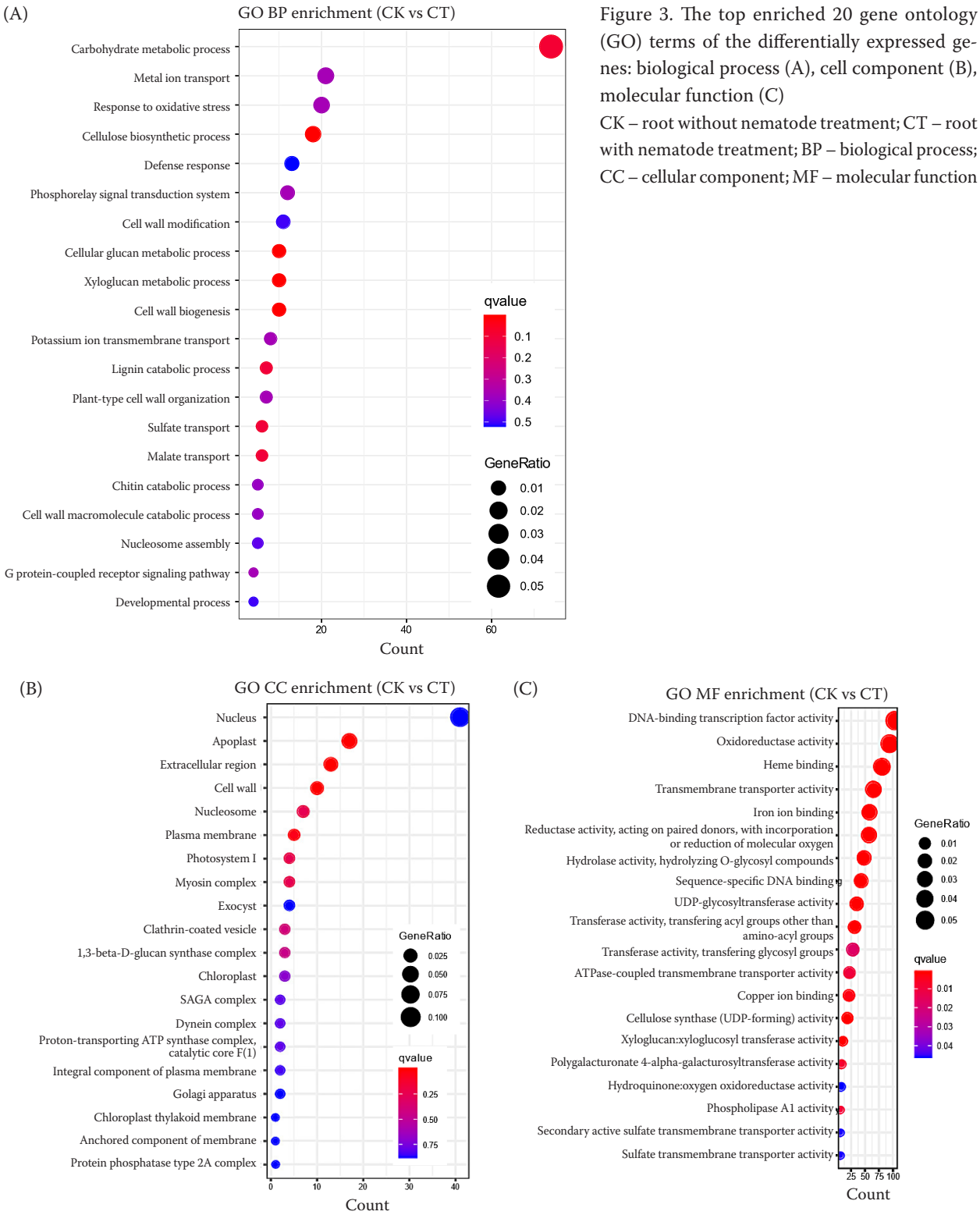

Figure 3. The top enriched 20 gene ontology (GO) terms of the differentially expressed genes: biological process (A), cell component (B), molecular function (C)

CK – root without nematode treatment; CT – root with nematode treatment; BP – biological process; CC – cellular component; MF – molecular function

DISCUSSION

Compared with roots of peach and Mirabolano 29C after a 60-day treatment of root-knot nematodes, we found that there was no difference between the roots that were grown in normal conditions and those treated with nematodes. This can suggest that Mirabolano 29C does show resistance to root-knot nematodes. However, when faced with nematodes, the roots of the peach appeared swollen and grey,

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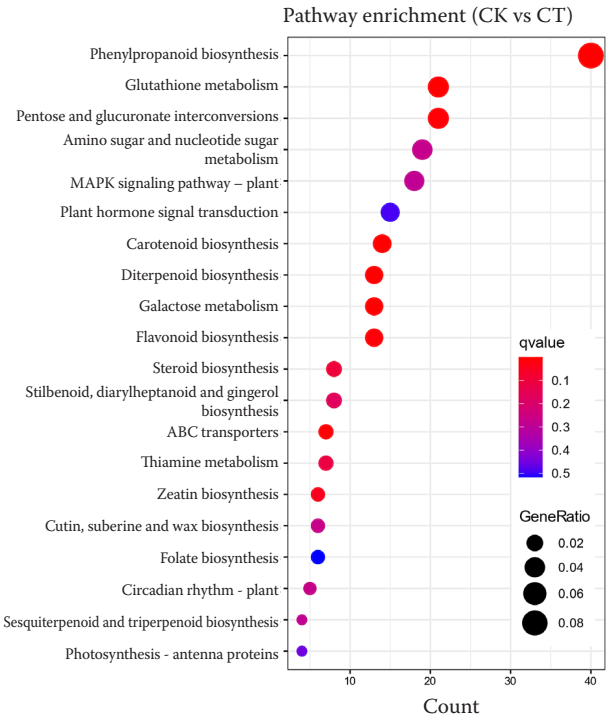

Figure 4. The top enriched 20 Kyoto Encyclopedia of Genes and Genomes (KEGG) pathways of the differentially expressed genes

CK – root without nematode treatment; CT – root with nematode treatment

and the fibrous roots were reduced. The results indicated that Mirabolano 29C indeed showed stronger resistance to root-knot nematodes. Transcriptome sequencing is a very simple, but effective, way that can give us some clues why Mirabolano 29C has stronger resistance, or which genes increase its resistance.

In this study, we tried to explore some key genes that may play important roles in root-knot nematodes resistance of Mirabolano 29C. After comparing the RNA-seq results of two groups (grown under normal conditions and treated with nematodes for 60 days), 3 176 genes were differentially expressed and there were less up-regulation genes than the down-regulation genes. Through the KEGG enrichment analysis, the DEGs were found to be significantly enriched into the "Phenylpropanoid biosynthesis" pathway. In the evolution of plants, secondary metabolites are considered to play a key role in the defence against pathogen invasions, and phenylpropane is a major player (Weisshaar & Jenkins 1998; Vogt 2010; Bauters et al. 2021). For example, a study in rice *(Oryza sativa* L.) showed that the phenylpropanoid pathway will stimulate the systemic defence activation by cationic chitosan oligomers with anionic pectin oligomers COS-OGA against root-knot nematodes (Singh et al. 2019). Another crucial secondary metabolite is plant flavonoids which will be induced during a nematode infection in the plant roots which play a role in the defence responses against a variety of microorganisms (Schmid et al. 1990; Lu et al. 2017; Hamamouch et al. 2020). In addition, glutathione metabolism was also considered to have contributions to plant resistance against root-knot nematodes (Meher et al. 2011). In our study, DEGs were significantly enriched into the flavonoid biosynthesis and glutathione metabolism pathways. These results indicate that the genes that may improve the root resistance to root-knot nematodes quickly responding to the environmental stress leading to a tolerance to root-knot nematodes in Mirabolano 29C.

The phenylpropanoid pathway is also responsible for the biosynthesis of some related polymers, including lignin (Vogt 2010). Furthermore, lignin, which has been proven to provide resistance against pathogens, is a plant biopolymer that presents in the cell walls (Fagerstedt et al. 2010). When the cell wall is damaged, the lignin deposition can reinforce it to maintain functional integrity (Denness et al. 2011). In this study, we can find that the DEGs were enriched into the cell wall biogenesis. The cell wall of plants is a complex and dynamic structure. Its functional integrity will be monitored and maintained constantly during a plant's development and its interactions with the environment (Denness et al. 2011). Thus, we can speculate that the cell wall damage due to root-knot nematode infestation may be reduced due to the lignin synthesis in the phenylpropane pathway. Of course, the anti-root knot nematode effect of Mirabolano 29C is not a single regulatory pathway and may involve the joint action of many molecules. For example, in the GO enrichment analysis, we found that the DEGs were also enriched in other signalling pathways closely related to the stress response, such as the response to oxidative stress, defence response, and trehalose biosynthetic process. These results can provide a theoretical reference for the selection and breeding of rootstocks for disease and insect resistance.

CONCLUSION

In this study, peach seedlings and plum (Mirabolano 29C) seedlings grown for 1 year under normal conditions were simultaneously infected with root-

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knot nematodes for 60 days and compared with roots under normal growth for 60 days. It was found that the root system of Mirabolano 29C showed no significant difference between the nematode-treated and untreated conditions. However, the roots of the commonly used peach rootstocks showed multiple swellings and reduced fibrous roots after infestation with nematodes. This proves that Mirabolano 29C is more resistant to a root-knot nematode infection. Next, we compared the root systems of the root-knot nematode-treated and normally grown Mirabolano 29C by transcriptome sequencing and found that more than 3 000 differentially expressed genes were generated between the two groups. These differentially expressed genes were enriched into the phenylpropanoid biosynthesis and flavonoid biosynthesis pathways. These results suggest that phenylpropane and flavonoids may play a key role in the resistance of Mirabolano 29C to root-knot nematodes, but how they regulate the nematode resistance of plants still needs to be further explored. This study is the first to report the information related to root-knot nematode resistance of rootstocks, which can provide an important reference value for root-knot nematode resistance and disease resistance breeding of other rootstocks.

Data availability. The sequencing reads were deposited in the Sequence Read Archive (SRA) of the GenBank (https://www.ncbi.nlm.nih.gov/genbank) under the BioProject No. PRJNA809767.

REFERENCE

Ashburner M., Ball C.A., Blake J.A., Botstein D., Cherry J.M. (2000): Gene ontology: Tool for the unification of biology. The gene ontology consortium. Nature Genetics, 25: 25–29.

Bauters L., Stojilković B., Gheysen G. (2021): Pathogens pulling the strings: Effectors manipulating salicylic acid and phenylpropanoid biosynthesis in plants. Molecular Plant Pathology, 22: 1436–1448.

Birwal P., Deshmukh G., Saurabh S., Pragati S. (2017): Plums: A brief introduction. Journal of Food, Nutrition and Population Health, 1: 1–5.

Byrne D.H., Raseira M.B., Bassi D., Piagnani M.C., Gasic K., Reighard G.L., Moreno M.A., Pérez S. (2012): Peach. In: Badenes M.L., Byrne D.H. (eds.): Fruit Breeding. Springer: 505–569.

Chen Y., Chen Y., Shi C., Huang Z., Zhang Y., Li S., Li Y., Ye J., Yu C., Li Z. (2018): SOAPnuke: A MapReduce acceleration-supported software for integrated quality control and preprocessing of high-throughput sequencing data. Gigascience, 7: gix120.

Denness L., McKenna J.F., Segonzac C., Wormit A., Madhou P., Bennett M., Mansfield J., Zipfel C., Hamann T. (2011): Cell wall damage-induced lignin biosynthesis is regulated by a reactive oxygen species-and jasmonic acid-dependent process in Arabidopsis. Plant Physiology, 156: 1364–1374.

Dodds P.N., Rathjen J.P. (2010): Plant immunity: Towards an integrated view of plant–pathogen interactions. Nature Reviews Genetics, 11: 539–548.

Fagerstedt K.V., Kukkola E.M., Koistinen V.V., Takahashi J., Marjamaa K. (2010): Cell wall lignin is polymerised by class III secretable plant peroxidases in Norway spruce. Journal of Integrative Plant Biology, 52: 186–194.

Faust M., Suranyi D., Nyujto F. (1998): Origin and dissemination of apricot. In: Janick J. (ed.): Horticultural Reviews. Vol. 22, Westport, New York, John Wiley & Sons, Inc.: 225–260.

Forghani F., Hajihassani A. (2020): Recent advances in the development of environmentally benign treatments to control root-knot nematodes. Frontiers in Plant Science, 11: 1125.

Guo Z., Tang G., Zhou Y., Shuwu L., Hou H., Chen Z., Chen J., Hu C., Wang F., De Smedt S.C. (2017): Fabrication of sustained-release CA-PU coaxial electrospun fiber membranes for plant grafting application. Carbohydrate Polymers, 169: 198–205.

Hamamouch N., Winkel B.S., Li C., Davis E.L. (2020): Modulation of *Arabidopsis* flavonol biosynthesis genes by cyst and root-knot nematodes. Plants, 9: 253.

Kanehisa M., Goto S., Kawashima S., Okuno Y., Hattori M. (2004): The KEGG resource for deciphering the genome. Nucleic Acids Research, 32 (Database Issue): D277.

King S.R., Davis A.R., Liu W., Levi A. (2008): Grafting for disease resistance. HortScience, 43: 1673–1676.

Li B., Dewey C.N. (2011): RSEM: Accurate transcript quantification from RNA-Seq data with or without a reference genome. BMC Bioinformatics, 12: 323.

Love M.I., Huber W., Anders S. (2014): Moderated estimation of fold change and dispersion for RNA-seq data with DESeq2. Genome Biology, 15: 1–21.

Lu Y., Chen Q., Bu Y., Luo R., Hao S., Zhang J., Tian J., Yao Y. (2017): Flavonoid accumulation plays an important role in the rust resistance of *Malus* plant leaves. Frontiers in Plant Science, 8: 1286.

Lu Z.-X., Sosinski B., Reighard G., Baird W., Abbott A. (1998): Construction of a genetic linkage map and identification of AFLP markers for resistance to root-knot nematodes in peach rootstocks. Genome, 41: 199–207.

https://doi.org/10.17221/111/2022-CJGPB

Meher H.C., Gajbhiye V.T., Singh G. (2011): Salicylic acid-induced glutathione status in tomato crop and resistance to root-knot nematode, *Meloidogyne incognita* (Kofoid & White) Chitwood. Journal of Xenobiotics, 1: e5-e5.

Melnyk C.W., Meyerowitz E.M. (2015): Plant grafting. Current Biology, 25: R183–R188.

Notaguchi M., Kurotani K.-I., Sato Y., Tabata R., Kawakatsu Y., Okayasu K., Sawai Y., Okada R., Asahina M., Ichihashi Y. (2020): Cell-cell adhesion in plant grafting is facilitated by β-1, 4-glucanases. Science, 369: 698–702.

Opperman C.H., Taylor C.G., Conkling M.A. (1994): Root-knot nematode – directed expression of a plant root – specific gene. Science, 263: 221–223.

Patterson J., Carpenter E.J., Zhu Z., An D., Liang X., Geng C., Drmanac R., Wong G.K.-S. (2019): Impact of sequencing depth and technology on de novo RNA-Seq assembly. BMC Genomics, 20: 1–14.

Pertea M., Kim D., Pertea G.M., Leek J.T., Salzberg S.L. (2016): Transcript-level expression analysis of RNA-seq experiments with HISAT, StringTie and Ballgown. Nature Protocols, 11: 1650–1667.

Rao J., Peng L., Liang X., Jiang H., Geng C., Zhao X., Liu X., Fan G., Chen F., Mu F. (2020): Performance of copy number variants detection based on whole-genome sequencing by DNBSEQ platforms. BMC Bioinformatics, 21: 1–14.

Rio D.C., Ares M., Hannon G.J., Nilsen T.W. (2010): Purification of RNA using TRIzol (TRI reagent). Cold Spring Harbor Protocols, 2010: pdb. prot5439.

Schmid J., Doerner P.W., Clouse S.D., Dixon R.A., Lamb C.J. (1990): Developmental and environmental regulation of a bean chalcone synthase promoter in transgenic tobacco. The Plant Cell, 2: 619–631.

Singh R.R., Chinnasri B., De Smet L., Haeck A., Demeestere K., Van Cutsem P., Van Aubel G., Gheysen G., Kyndt T. (2019): Systemic defense activation by COS-OGA in rice against root-knot nematodes depends on stimulation of the phenylpropanoid pathway. Plant Physiology and Biochemistry, 142: 202–210.

Thomas H.R., Frank M.H. (2019): Connecting the pieces: Uncovering the molecular basis for long-distance communication through plant grafting. New Phytologist, 223: 582–589.

Vogt T. (2010): Phenylpropanoid biosynthesis. Molecular Plant, 3: 2–20.

Weisshaar B., Jenkins G.I. (1998): Phenylpropanoid biosynthesis and its regulation. Current Opinion in Plant Biology, 1: 251–257.

Wickham H. (2011): ggplot2. Wiley Interdisciplinary Reviews: Computational Statistics, 3: 180–185.

Xu G., Yuan M., Ai C., Liu L., Zhuang E., Karapetyan S., Wang S., Dong X. (2017): uORF-mediated translation allows engineered plant disease resistance without fitness costs. Nature, 545: 491–494.

Xu X., Fang P., Zhang H., Chi C., Song L., Xia X., Shi K., Zhou Y., Zhou J., Yu J. (2019): Strigolactones positively regulate defense against root-knot nematodes in tomato. Journal of Experimental Botany, 70: 1325–1337.

Young M.D., Wakefield M.J., Smyth G.K., Oshlack A. (2010): Gene ontology analysis for RNA-seq: accounting for selection bias. Genome Biology, 11: 1–12.

Yu G., Wang L.-G., Han Y., He Q.-Y. (2012): clusterProfiler: An R package for comparing biological themes among gene clusters. Omics, 16: 284–287.

Zhang G., Guo G., Hu X., Zhang Y., Li Q., Li R., Zhuang R., Lu Z., He Z., Fang X. (2010): Deep RNA sequencing at single base-pair resolution reveals high complexity of the rice transcriptome. Genome Research, 20: 646–654.

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