University of Zabol Faculty of Agriculture Department of Plant Protection

The Thesis Submitted for the Degree of M. Sc (in the Field of Plant Pathology)

Genetic diversity of anamorph of Venturia inaequalis in khorasak Razavi and Shomali khorasan provinces

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Abstract

Apple scab is the most destructive disease of apple worldwide. Generally, damage of apple scabe is more than other diseases of apple. It includes: loss of yield, quality of fruit, damage of branch and leaf of trees, and increase of gardener expenses in terms of fungicide application. This research was performed to evaluate the genetic diversity Fusicladium pomi (anamorph of Venturia inaqualis) isolates in Khorasan Razavi and Shomali Khorasan provinecs using molecular markers as rep-PCR and RAPD PCR. Sampling was done from apple garden of Shirvan, Bojnoord, Esfarayen, Ohochan, Dargaz, Torghabeh and Shandize during summer 2014 sampled. Samples (Leaf and fruit) were transfer to laboratory and were cultured on Potato Dextrose Agar and Water Agar medium. All fungal growth were sub cultured followed purified on WA and then were identified using valid key. Of 300 fungal isolated, 70 isolate were identified as V. inaqualis. The genetic diversity of 26 selected isolates from different region was assayed using two above marker. DNA extraction was carry out using CTAB procedure. Amplification of expects fragment was done using 23 RAPD primers. The result of pcr amplication showed that from 23 primer 12 primers were able to amplify respected fragments. All three used primers in Rep assay were able to produce corresponding fragments. REP primers amplified a total number of 40 bands at range of 240 to 3000 bais pair. RAPD primers also amplified a total of 118 bands at range of 250 to 2500 bp. Matrix of similarity between isolates was calculated based on Jaccard coefficient and clustering analysis and dendrogram design was done based on UPGMA using NTSYSPC 2.10 software. The results of rep-PCR analysis showed that isolate in similarity level 66 % divided to 14 groups. the highest variation was observed for Ghochan isolates and the lowest was related to Esfarayen isolates. Disease severity assay showed that there is a direct relation between disease severity and rate of genetic variation. The funding of this work indicate that the rate of genetic variation of F. pomi is high in different region of Khorasan Razavi and Shomali Khorasan provinecs. Based on our results, we could conclude that Rep marker is power than RAPD marker and is a suitable marker to determine genetic diversity of this fungus

Keywords: apple scabe disease; genetic diversity; molecular markers.