J. Turk. Phytopath., Vol. 46 No. 2, 61-67, 2017 ISSN 0378 - 8024

Molecular Characterization of *Rhizoctonia* Species and Anastomosis Groups in Barley Production Areas in Ankara Province

Filiz ÜNAL[1] M. Erdi KARA[2]

[1] Plant Protection Central Research Institute, Ankara
[2]Ankara University, Faculty of Agriculture, Department of Field Crops, Ankara
Corresponding author email: fucar06@yahoo.com

Accepted by 14 December 2017

ABSTRACT

Barley (*Hordeum vulgare* L.) is an important cereal crop in terms of human and animal nutrition in the world. *Rhizoctonia* species are a diverse group of fungi that damage small-grain cereals by rotting the seedlings, roots and crowns. In order to determine the root rot diseases, surveys were carried out in barley fields in 2015 growing season in Ankara. 48 barley and 48 soil samples were collected from barley fields. As a result of isolation from plant and soil samples collected from barley fields, 28 *Rhizoctonia* isolates belonging to 9 anastomosis groups were obtained. The isolates were identified based on hyphal, colony morphology, anastomosis reaction with known tester isolates and rDNA-ITS sequence analysis. Multinucleate (MN) *Rhizoctonia* isolates were grouped into five AGs as *R. solani* AG 2-2, AG 4 HG II, AG 5, *Waitea circinata* var. *circinata*, *Waitea circinata* var. oryzae and Binucleate (BN) *Rhizoctonia* isolates were grouped into four AGs as AG D I (*R. cerealis*), AG A, AG E and AG I. As a result of pathogenicity tests, AG 2-2, AG 4 HG II, AG 5, *Waitea circinata* var. *circinata, Waitea circinata* var. *oryzae* and BN AG D I (*R. cerealis*) groups were found to be pathogen on barley and it was determined that the most virulent group was AG 4 HG II.

Key words: *Rhizoctonia* spp., anastomosis group, barley, rDNA-ITS sequence

ÖZET

Ankara İli Arpa Üretim Alanlarındaki *Rhizoctonia* Türlerinin Anastomosis Gruplarının Moleküler Karakterizasyonu

Arpa (*Hordeum vulgare* L.), dünyada insan ve hayvan beslemesi açısından önemli bir hububat bitkisidir. *Rhizoctonia* türleri hububatta fide çürüklüğü, kök ve kökboğazı çürüklüğü yaparak zarar oluşturan kendi içinde değişik gruplara sahip funguslardır. 2015 üretim sezonunda kök çürüklüğü hastalıklarını belirlemek için Ankara İli arpa tarlalarında surveyler gerçekleştirilmiştir. Bu tarlalardan 48 bitki ve 48 toprak örneği toplanmıştır. Bu bitki ve toprak örneklerden yapılan izolasyonlar sonucunda 9 anastomosis grubuna ait 28 Rhizoctonia izolatı elde edilmiştir. İzolatlar hif yapısı, koloni morfolojisi, test izolatları ile anastomosis reaksiyonları ve rDNA-ITS sekans analizleri ile tespit edilmiştir. Çok çekirdekli *Rhizoctonia* izolatları *R. solani* AG 2-2, AG 4 HG II, AG 5, *Waitea circinata* var. *circinata* ve *Waitea circinata* var. *oryzae* olarak beş grup içerisinde yer almıştır. İki çekirdekli *Rhizoctonia* izolatları *R. solani* AG D I (*R. cerealis*), AG A, AG E ve AG I.olarak dört grup içerisinde yer almıştır. Patojenite testleri sonucunda AG 2-2, AG 4 HG II, AG 5, *Waitea circinata* var. *circinata, Waitea circinata* var. *oryzae* ve BN AG D I (*R. cerealis*) grupları arpada patojen bulunmuştur. En virulent grup AG 4 HG II olarak tespit edilmiştir.

Anahtar Sözcükler: *Rhizoctonia* spp., anastomosis grup, arpa, rDNA-ITS sekans

INTRODUCTION

Barley (*Hordeum vulgare* L.) is one of the important crop of agriculture, and it is the second most important cereal grain with 6,7 million tone production in Turkey. It is used as malt in brewing and distilling industry, as an additive for animal feed, and as a component of various food and bread for human consumption. Ankara Province is the second place in barley production after Konya Province in Central Anatolia Region.

Root and crown rots are important and widespread cereal diseases that are present in most parts of Turkey, including the Central Anatolia Region. Root and crown or foot rot is considered a disease complex. One of the main root and crown rot pathogens on wheat (*Triticum* spp.*)* and barley (*Hordeum vulgare* L.) are *Rhizoctonia* species. The genus *Rhizoctonia* include a wide variety of genetically diverse fungi associated with plant roots and soil that are economically important plant pathogens, saprophytes on organic matter, and symbionts associated with orchids, mosses and liverworts (Gonzales et al. 2006). They are composed of several anastomosis group. 14 Anastomosis groups (AGs) (AGs 1-13 and BI) and 12 subgroups (AGs 1-IA, 1-IB, 1-IC, 2-1, 2-2IIB, 2-2IV, 4HG-I, 4HG-II, 6HG-I, 6GV, 9TP, 9TX) have been identified on the basis of hyphal anastomosis interactions, cultural and morphological characters, host range, pathogenicity, biochemical reactions and molecular studying in *R. solani. R. zeae and R. oryzae* have been assigned to WAG-Z and WAG-O, respectively (Sneh et al. 1996). Binucleate *Rhizoctonia* isolates are grouped into 16 AGs (AG A, B, C, D, E, F, G, H, I, K, L, O, P, Q, R, S (Sharon et al. 2008).

Barley is sensitive to infection by several anastomosis groups (AGs) and subgroups of *Rhizoctonia*. *R. solani* AG 1-IB, AG 2-1, AG 2-2, AG 3, AG 4 HG-II, AG 5, AG 8, AG 9, AG 11, *Waitea circinata* var. *circinata, Waitea circinata* var. *zeae* and *Waitea circinata* var. *oryzae,* binucleate AG A, AG C, AG D (*Rhizoctonia cerealis*), AG E, AG H, AG I and AG K were determined on barley in the world (Li and Xia 1988, Xia and Li, 1989, Ogoshi et al. 1990, Tewoldemedhin et al. 2006, Tomaso-Peterson ve Trevathan 2007, Schroeder et al. 2011). *Rhizoctonia solani* AG 2-1, 3, 4, 5, AG 11, *Waitea circinata* have been reported previously to be pathogenic and AG K has been reported to be nonpathogenic on barley in Erzurum, Turkey (Demirci, 1998). *Rhizoctonia* species cause root and crown rot, damping off and stunting on barley.

Various molecular markers have been used for characterization and grouping *Rhizoctonia* species. The genetic diversity of *Rhizoctonia* isolates has been studied using RAPD-PCR, SSR-PCR, rDNA-RFLP, rDNA-ITS sequence analysis, universally primed-PCR and rep-PCR (Sharon et al. 2006). Currently, the rDNA-ITS sequence analysis is the most appropriate method for classification of *Rhizoctonia* spp and sequence analysis of the ITS-5.8S rDNA region has been used as a suitable molecular tool for identification of *R. solani* subgroups (Carling et al. 2002; Hyakumachi et al. 1998; Priyatmojo et al. 2001; Salazar et al. 2000).

The objectives of this study were to identify the species of pathogenic and nonpathogenic *Rhizoctonia* and subgroups causing root and crown rot and stunting on barley in Ankara Province

MATERIALS and METHODS

Plant collection and isolation

In order to determine the anastomosis groups and pathogenicity of *Rhizoctonia* species associated with stunting, bare patch, root and crown rot of barley in Ankara, 48 samples of barley and 48 rhizosphere soil were collected in 2015 growing season. Plant and soil samples were taken from fields in five districts [Polatlı (20 plant-20 soil), Gölbaşı (8-8), Bala (5-5), Haymana (4-4), Beypazari (11-11)] of Ankara Provinces. Segments of necrosed tissue were placed on acidified water agar (1.5 % water agar amended 3 ml of 10 % lactic acid per liter of medium). Hyphal tips of *Rhizoctonia*-like fungi were transferred to Potato Dextrose Agar (PDA, Merck, Germany) containing 50 mg/l streptomycin sulfate.

Sterile barley straws were used for *Rhizoctonia* spp. isolation from soil samples. Soil samples (collected with plants) from the respective fields were transferred to pots on a greenhouse bench (20±20C). Pots were then watered to field capacity about 4 cm long internodal segments of mature, dried barley straw were inserted vertically, 4 per pot, and left for 3 or 4 days After that straws were removed, washed, blotted and placed on acidified water agar. Isolates of *Rhizoctonia* were transferred to PDA medium (Ogoshi et al. 1990).

Identification

In order to determine hyphal diameter and the number of nuclei per cell of the isolates, *Rhizoctonia* isolates were maintained on PDA in an incubator regulated at 25 °C and in darkness. Developing mycelia were stained with safranin O (Sigma, USA) and 3% KOH (Bandoni, 1979) and observed under phase contrast microscopy at x 400 magnification. Hyphal diameter was determined by measuring 10 cells. Nuclei were counted in 15 cells. Anastomosis was tested by pairing isolates with representative testers of *Rhizoctonia* spp. isolates were activated on PDA at 25 °C in the dark. Coverslips, sterilized by dipping in 95 % ethyl alcohol and flaming, were coated with a thin layer of 0.5 % PDA and placed on water agar plates. Agar plugs with mycelia of *Rhizoctonia* isolates and the tester isolates were cut the margin of a growing colony and transfered to water agar plates on the opposite sides of the coverslip. After incubation at 25 °C for 24-48 h in the dark, when overlapping mycelia of two isolates were observed, the coverslip was removed from the plate and placed on a slide in the mixture of one drop of safranin O and one drop of 3 % KOH. Stained hyphae were observed microscopically. Anastomosing hyphae were traced back to their source in order to confirm the anastomosis between our isolates and the tester isolates (Kronland and Stanghellini, 1988). For the anastomosis testing, all pairs were examined twice.

Molecular Analysis

Approximately, 300 mg mycelium were harvested and ground with liquid nitrogen in a sterile mortar for DNA extraction from culture medium. Genomic DNA was extracted using a Qiagen DNeasy ® Plant Mini Kit, as specified by the manufacturer, and stored at -20 0C prior to use. PCR reaction mixtures and condition were modified from previous studies (Aroca and Raposo 2007; Cobos and Martin, 2008). The reaction mixtures of PCR, a final volume of 50 µl, contained 5µl of 10X buffer [75 mM Tris HCl, pH 9.0, 50 mM KCl, 20 mM (NH4)2SO4], 2 µl of 5 µM each primers, 5 µl of 1.5mM MgCl2, 2 µl of 10 mM deoxynucleoside triphosphates (dNTPs), 1 U Taq polymerase (Fermatas), 5 µl of DNA template for each reaction and 5 µl of bovine serum albümin (BSA: 10 mg/ml). DNA amplifications were carried out in a Techne TC-5000 thermal cycler by the following program: 94 C for 2 min, followed by 34 cycles of (1) denaturation (94˚C for 30 s), (2) annealing (60˚C for 30 s) and (3) extention (72 ˚C for 30 s), and a final extension step 10 min at 72˚C. The ITS region of the isolate was amplified using the universal primers ITS-1 (5’ TCC GTA GGT GAA CCT GCGG 3’) and ITS-4 (5’ TCC TCC GCT TAT TGA TATGC 3’). The PCR products were separated in 1.5 % agarose gels stained with ethidium bromide, and visualized under UV light. They were sequenced by GENOKS (Gene Research and Biotechnology Company, Ankara, Turkey).

Pathogenicity tests

First pathogenicity test was conducted with agar-plate assay with all isolates (Ichielevich-Auster et al. 1985). Pathogenicity was also tested on seedlings grown in pots the most virulent isolate on susceptible barley cultivar (cv.Kıral 97). Seedlings were grown in plastic pots (5 cm in diameter) in the greenhouse conditions at 23 ± 2 °C with a 12-h photoperiod and 50–60 % RH. Inoculum was prepared on the moistened sterile wheat grains in tests tubes. The bottom of the pots were filled with 40 cm^3 of medium vermiculite and topped with 30 cm^3 of natural sterile silt loam. Eight pathogen-colonized wheat grains were placed in the middle of the soil layer. Control was consisted of pots without inoculum. All pots were covered with clear polyethylene and incubated 5 days. There were six replicate pots for treatment. After 5 days, eight seeds of barley were placed on the soil surface, covered with 10 cm^3 of sterile natural top soil and watered with 10 ml of distilled water. After 3 weeks, the plants were washed free of soil (Paulitz et al. 2003). Severity of *Rhizoctonia* root rot was evaluated on a scale of 0 to 8 (Kim et al. 1997).

RESULTS AND DISCUSSION

Ninety six plant and soil samples were collected from the barley growing areas showing stunting, root and crown rot in Ankara Province and total of 28 *Rhizoctonia* isolates (*R. solani* AG 2, 4 HG II, 5, *Waitea circinata* var. *circinata, Waitea cir.* var. *oryzae*, Binucleate AG DI, A, E and I) were obtained (Figure 1). Fifteen isolates from plants and 13 isolates from soil (Table 1) were identified with the help of their vegetative hyphae, nuclear condition, hyphal anastomosis reaction with known tester isolates. C2 and C3 anastomosis reactions were observed between *Rhizoctonia* isolates and test isolates diagnosed in anastomosis reaction studies (Figure 2). Isolates were also identified by using rDNA-ITS sequence analysis for supporting hyphal anastomosis reactions and determining further subgroups. The resulting sequences were compared to other *Rhizoctonia* sequences and were 98 to 100% identical to other *Rhizoctonia* sequences in the GenBank. In the studies that were performed in different regions in world, it was determined that there were different anastomosis groups pathogen and non-pathogen disease on barley and they showed similarity with the groups that were determined in our study. (Li and Xia 1988, Xia and Li, 1989, Ogoshi et al. 1990, Tewoldemedhin et al. 2006, Tomaso-Peterson ve Trevathan 2007, Schroeder et al. 2011). Previously, a study was carried out in Turkey for determining anastomosis groups in barley. *R. solani* AG 2-1, AG 3, AG 4, AG 5, AG 11, *Waitea circinata var. circinata* and binucleate AG K were determined by Demirci in Erzurum province (Demirci, 1998). *R. solani* AG 2-2, *Waitea circinata var. oryzae,* Binucleate AG A, DI, I and E groups were determined in this study different from the study performed by Demirci, 1998.

Table 1. Diseases severity and number of isolates obtained from *Rhizoctonia* species and anastomosis groups isolated from plant and soil samples in Ankara Districts

***Rhizoctonia* species**	**Anastomosis groups**	**Origin (District)**	**Host (plant or soil)**	**Number of isolate**	**Diseases severity(%)**
Rhizoctonia solani	AG 2-2	Polatlı	plant	1	72
	AG 4 HG II	Polatlı, Beypazarı, Gölbaşı	plant	5	88-98
	AG 5	Polatlı, Bala, Haymana, Gölbaşı	plant and soil	4	70-85
Waitea circinata	var. *circinata*	Polatlı, Beypazarı	plant and soil	3	78-85
	var. *oryzae*	Polatlı, Gölbaşı	plant	2	85-90
Binucleate *Rhizoctonia* spp	AG DI (*R. cerealis*)	Polatlı, Haymana, Beypazarı	plant and soil	4	78-86
	AG A	Bala, Haymana	soil	3	0
	AG E	Polatlı, Bala	soil	2	0
	AG I	Polatlı, Gölbaşı, Beypazarı	plant and soil	4	0

While MN isolates were most obtained from the plant, BN isolates were generally isolated from the soil in the study that was carried out similarly by Juan-Abgona et. al (1996) in Japan and Chen and Chuang (1997) in Taiwan. Of the isolates obtained, 53,58% were MN isolates and 46.42% were BN isolates.

Regarding the pathogenicity test on agar plates and greenhouse conditions, AG 2-2, AG 4 HG II, AG 5, *Waitea circinata* var. *circinata, Waitea circinata* var. *oryzae* and BN AG D I (*R. cerealis*) groups were found to be pathogenic on barley (Table 1). *Rhizoctonia* isolates caused significant reduction of emergence, stunting, reduction in the number of seminal roots and superficial discolouration on the hypocotyls and roots on barley in the pathogenicity tests. *Rhizoctonia solani* AG 2-1, 3, 4, 5, AG 11, *Waitea circinata* have been reported previously to be pathogenic and AG K has been reported to be nonpathogenic on barley in Erzurum, Turkey (Demirci, 1998). In present study, all binucleate isolates except for *R. cerealis* AG D I were not pathogen on barley. When the studies carried out in the world were examined, nonpathogenic BN species was found in the barley except for AG D. Although there are few studies reporting that binucleate *Rhizoctonia* spp. pathogen on some other hosts, these species generally show saprophytic character in the soil. Among them, low-virulent or non-virulent species show hypo-virulent property (Sneh et al. 1996; Tewoldemedhin et al. 2006). These groups, generally composed of binucleate species, may also be used for the biological control studies (Cardoso and Echandi, 1987 a,b; Roberts and Sivasithamparam 1986; Gutierrez and Torres 1990; Herr 1995).

Figure 1. Colony appearance on PDA of some of the obtained *Rhizoctonia* isolates
(a) AG 2, (b) AG 4, (c) *Waitea circinata* var. *circinata,* (d) AG I

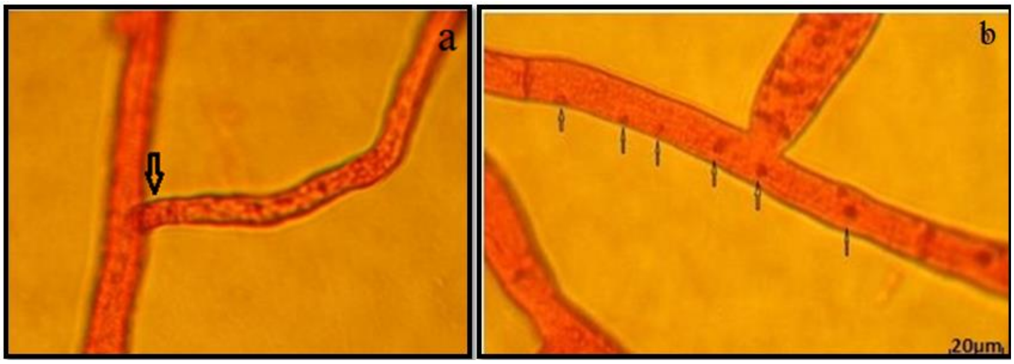

Figure 2. C3 anastomosis reaction between hyphae (a) and multinucleate hyphae cell (b)

In this study, all isolates belonging to *R. solani* AG 4 HG II group that was isolated from three different districts constituted the most virulent group by causing dark brown and severe lesions in the root and hypocotyls. Previous studies shown that AG 4 was the most virulent group. Results of these studies are parallel with our findings. This group has high virulent characteristic at the same way in many cultivated plants (tomato, potato, melon, broccoli, spinach, beans, soya bean, onion, pine seed, peas, cotton and peanut) (Kural et al. 1994, Sneh et al. 1996, Demirci 1998, Yıldız and Döken 2002, Eken and Demirci 2004, Tewoldemedhin et al. 2006, Tomaso-Peterson and Trevathan 2007).

Rhizoctonia is one of the main causal agents of dryland root rot on wheat and barley in Turkey. It is known that the host range and diseases caused by the isolates from different species or anastomosis group are different. Therefore, determination of the species or anastomosis grouping of the isolates is very important. Results of this study present the first report of *Rhizoctonia* species, causing disease of barley in Central Anatolia Region. Due to the severe symptoms, it should be considered a potential threat to barley cultivation in Central Anatolia Region, Turkey.

LİTERATÜR LİSTESİ

Aroca, A., Raposo, R**.** 2007. PCR-based stragety to detect and identify species of *Phaeoacremonium* causing grapevine diseases. Applied and Environmental Microbiology, 73, 2911-2918.

Bandoni, R.J. 1979. Safranin O as a rapid stain for fungi. Mycologia 71:873–874.

Cobos, R., Martin, M.T. 2008. Molecular characterisation of *Phaeomoniella chlamydospora* isolated from grapevines in Castilla y León (Spain). Phytopathol. Mediterr., 47, 20–27.

Demirci, E. 1998. *Rhizoctonia* species and anastomosis groups isolated from barley and wheat in Erzurum, Turkey, Plant Pathology, 47(1): 10-15.

González, D., Cubeta M.A., Vilgalys R. 2006. Phylogenetic utility of indels within ribosomal DNA and _-tubulin sequences from fungi in the *Rhizoctonia solani* species complex. Molecular Phylogenetics and Evolution 40 (2006) 459–470.

Sharon, M., Kuninaga, S., Hyakumachi, M., Naito, S. and Sneh, B. 2008. Classification of *Rhizoctonia spp*. using rDNA-ITS sequence analysissupports the genetic basis of the classical anastomosis grouping. Mycoscience, 49, 93–114.

Sneh, B., Jabaji-Hare, S., Neate, S. and Dijst, G., 1996. *Rhizoctonia* species: Taxonomy, molecular biology, ecology, pathology and diseases control. 1-559.

Tewoldemedhin, Y.T., Lamprecht, S.C., McLeod, A. and Mazzola, M. 2006. Characterization of *Rhizoctonia* spp. Recovered from crop plants used in rotational cropping systems in the Western Cape Province of South Africa. Plant Disease, 90(11): 1399-1406.

Kim, D.S., Cook, R. J., and Weller, D.M. 1997. *Bacillus* sp. L324-92 for biological control of three root diseases of wheat grown with reduced tillage. Phytopathology, 87: 551-558.

Li, Q.X. and Xia, Z.J. 1988. Study on the biological characters of *Rhizoctonia* isolated from some crops in Jiangsu. Journal of Jiangsu Agricultural College, 9(3): 23-26.

Xia, Z.J. and Li, Q.X. 1989. Preliminary study on aetiology of sharp eyespot in wheat and barley in Jiangsu, China. Acta Phytopathologica Sinica, 19(3): 135-139.

Tomaso-Peterson, M. and Trevathan, L.E. 2007. Characterization of *Rhizoctonia*-like fungi isolated from agronomic crops and turfgrasses in Mississippi. Plant Disease, 91(3): 260-265.

Ogoshi, A., Cook, R.J. and Bassett, E.N. 1990. *Rhizoctonia* species and anastomosis groups causing root rot of wheat and barley in the Pacific Northwest. Phytopathology, 80(9),785-788.

Schroeder, K.L., Shetty, K.K. and Paulitz, T.C. 2011. Survey of *Rhizoctonia* spp. from wheat soils in the U.S. and determination of pathogenicity on wheat and barley. Phytopathology, 101: 161s.

Carling, D.E., Kuninaga, S. and Brainard, K.A. 2002. Hyphal anastomosis reactions, rDNA-internal transcribed spacer sequences, and virulence levels among subsets of *Rhizoctonia solani* anastomosis group-2 (AG-2) and AG-BI. Phytopathology 92:43–50.

Hyakumachi, M., Priyatmojo, A., Kubota, M. and Fukui, H. 2005. New Anastomosis Groups, AG-T and AG-U,of Binucleate *Rhizoctonia* spp. Causing Root and Stem Rot of Cut-Flower and Miniature Roses. Phytopathology, 95(7): 784-792.

Priyatmojo, A., Yamauchi, R., Kageyama, K. and Hyakumachi, M. 2001. Grouping of Binucleate *Rhizoctonia* Anastomosis Group D (AG-D) Isolates into Subgroups I and II Based on Whole-Cell Fatty Acid Compositions. J. Phytopathology, 149, 421- 426.

Salazar, O., Julian M.C., Rubio V. 2000. Primer based on specific rDNA-ITS sequences for PCR detection of *Rhizoctonia solani*, *R. solani* AG 2 subgroups and ecological types, and binucleate *Rhizoctonia*. Mycol Res 104:281–285

Kronland, W.C., Stanghellini, M.E. 1988. Clean slide technique for the observation of anastomosis and nuclear condition of *Rhizoctonia solani*. Phytopathology 78:820–822.

Paulitz, T.C., Smith, J.D. and Kidwell, K.K. 2003. Virulence of *Rhizoctonia oryzae* on wheat and barley cultuvars from the Pacific Northwest. Plant disease, 87(1): 51-55.

Juan-Abgona, R.V., Katsuno, N., Kageyama, K., and Hyakumachi, M. 1996 Isolation and identification of hypovirulent *Rhizoctonia* spp. from soil. Plant Pathology, 45 (5): 896-904.

Ichielevich- Auster, M., Sneh, B., Koltin, Y. and Barash, I. 1985. Suppression of Damping-Off Caused by *Rhizoctonia* Species by a Nonpathogenic Isolate of *R. solani*. Phytopathology 75: 1080-1084.

Chen, C.S. and Chuang, T.Y. 1997. The anastomosis groups of binucleate *Rhizoctonia* in Taiwanese soils. Plant Pathology Bulletin, 6(4), 153-162.

Cardoso, J.E. and Echandi, E. 1987a. Biological control of *Rhizoctonia* root rot of snap bean with binucleate *Rhizoctonia*-like fungi. Plant Disease, 71:167-170.

Cardoso, J.E and Echandi, E. 1987b. Nature of protection of bean seedlings from *Rhizoctonia* root rot by a binucleate *Rhizoctonia*-like fungus, Phytopathology, 77, 1548-1551.

Gutiérrez, P. and Torres, H. 1990. Biological control of *Rhizoctonia solani* with binucleated *Rhizoctonia*. Fitopatología, 25, 2, 45-50 pp.

Roberts, F.A. and Sivasithamparam, K. 1986. Identity and pathogenicity of *Rhizoctonia* spp. associated with bare patch diseases of cereals at field site in Western Australia. Europan Journal of Plant Pathology, 92(5): 185-195.

Herr, L.J. 1988. Biocontrol of *Rhizoctonia* crown and root rot of sugar beet by binucleate *Rhizoctonia* spp. and *Laetisaria arvalis*, Annals of Applied Biology, 113,107-118.

Eken, C. and Demirci, E. 2004. Anastomosis groups and pathogenicity of *Rhizoctonia solani* and binucleate *Rhizoctonia* isolates from bean in Erzurum, Turkey. Journal of Plant Pathology 86(1): 49-52.

Kural, İ., Sagır, A. and Tatli, F. 1994. Characterization and Pathogenicity of Anastomosis Groups of *Rhizoctonia solani* İsolated from Cotton in Southeastern Turkey. 9. Congress of the Mediterranean Phytopathological Union. 18-24, Kuşadası Aydın, Türkiye. p. 117- 120.

Yıldız, A. and Döken, M.T. 2002. Anastomosis group determination of *Rhizoctonia solani* Kuhn (Telomorph: *Thanatephorus cucumeris*) isolates from tomatoes grown in Aydın, Turkey and their disease reaction on various tomato cultivars. Journal of Phytopathology -Phytopathologısch Zeithschrift, 150(10): 526-528.