Development of specific SCAR-markers for Meloidogyne incognita and Meloidogyne javanica

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ABSTRACT

Plant parasitic nematodes are major pests and cause diseases in many crops of economic importance. The annual losses world wide caused by plant parasitic nematodes are estimated to be approximately 77 billion US dollars. A substantial part of the losses is due to infestations by rootknot nematodes (Meloidogyne spp.) because they are parasites of a multitude of host plants. The most important and widely distributed root-knot nematode species are M. incognita, M. javanica, M. arenaria and M. hapla. For the development of specific sequence characterized amplified region (SCAR) markers from specific RAPD markers, samples of 20 populations of root knot nematodes were collected from heavily infested spots in the fields of nine different localities in four centres of Bani-Sweef governorate and 11 different localities in five centres of El Fayoum governorate. Such samples were inspected and the associated root knot nematodes species were isolated and identified depending upon the perennial patterns and two polymerase chain reaction (PCR) methods, which are ribosomal DNA (rDNA) and mitochondrial DNA (mtDNA). The random amplified polymorphic DNA (RAPD) method was used to investigate the genetic diversity in Meloidogyne species isolates. Populations of each of M. incognita and M. javanica were distinguished by differences in fragment patterns with any of the ten RAPD primers used. From analysis of RAPD fingerprints of all tested populations, two RAPD markers were detected, one was specific for M. incognita populations with primer OPK-2 at fragment size of 1000 bp and the second was specific for M. javanica populations with primer OPB-3 at size of 1100 bp. These two RAPD markers were converted into SCAR (specific characterized amplified region) markers, which were sequenced and two PCR primer pairs were designed for each of M. incognita (MIE-for and MIE-rev) and M. javanica (MJE-for and MJE-rev). These primers can be used to detect these nematode species in the fields.

Key words: SCAR-markers, specific PCR primers, plant parasitic nematodes.

INTRODUCTION

Poot-knot nematodes (RKN) constitute the most widely distributed group of plant-parasitic nematodes. These biotrophic endoparasites cause extensive damage to a wide variety of economically

important plants and are responsible for world wide annual loss, estimated at about 5% (Sasser and Carter, 1985).

Species identification in Meloidogyne has been a major component of taxonomic research in Nematology. Although there are approximately 60 described species of RKNs, most taxonomic attention has been focused on