



Molecular identification of three of the most important mealybug species (Hemiptera: Sternorrhyncha: Coccoidea: Pseudococcidae) on ornamental plants in Guilan province, Iran

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Abstract

Mealybugs (Coccoidea: Pseudococcidae) are serious pests, particularly as invasive species on many agricultural products. Morphological identification of mealybugs is based on adult female characters that, in the absence of adult females or with damaged specimens, can be problematic, especially when identification is required urgently, such as that involving the exportation/importation market. In this study, species-specific primers were designed to identify three of the most abundant mealybug species found on ornamental plants in Guilan province, Iran: *Planococcus citri* (Risso), *Pseudococcus viburni* (Signoret) and *Pseudococcus comstocki* (Kuwana). By generating amplification products of different sizes, the three species-specific primers, along with universal COI primers, were successfully used in multiplex PCR tests to identify all three mealybug species in a single reaction. Analysis of a large array of specimens from different geographic locations on different host plants showed that this was a reliable and accurate method.

Key words: PCR, taxonomy, Guilan province, Pseudococcidae

Introduction

Mealybugs (Hemiptera: Coccoidea: Pseudococcidae) are among the most destructive pests of many commercial crops worldwide (Miller *et al.* 2005). They can increase their population size rapidly (Franco *et al.* 2004). At large population levels, mealybugs can suck large amounts of nutrients from their hosts, severely weakening the plant. Also mealybugs secrete large amounts of honeydew on leaves and stems, providing a substrate on which sooty mould can grow and thus reduce the photosynthetic ability of the plant (Gullan *et al.* 2003). Moreover, in some cases, their ability to transmit viruses and other pathogens may damage the crop and, as a result, the relevant agricultural industry (Roivainen 1980; Bhat *et al.* 2005).

Mealybug species show a high degree of similarity in their morphology, particularly during the immature stages. Studies have shown that, under a variety of controlled environments, variation in factors such as temperature, relative humidity and host plant during their development can affect the morphology of the adult females, making them difficult to identify (Cox 2008). Because of their economic importance and the difficulties in identification, an accurate and fast identification method is needed, particularly one that does not rely on a specific life stage or gender. Such rapid and accurate identification is particularly important when the issue involves the exportation/importation of a commercial crop or for biological control studies, where accurate identification of the target species is vital because many parasitoids are species specific. For example, *Leptomastix dactylopii* Howard has been reported to be effective only against the citrus mealybug, *Planococcus citri* (Joyce *et al.* 2001; Daane *et al.* 2004; Walton & Pringle 2005), and therefore it is important to be sure that the host is *Pl. citri* before introduction. Molecular methods are becoming popular for the identification of eggs and immature stages (Armstrong & Ball 2005). There is no reliable method for morphological identification of immature mealybugs, except an identification key for six citrus mealybugs in Australia (Gullan 2000). Thus, a molecular approach would open up a new ave-