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ORIGINAL ARTICLE

Cantharidin content in two Mediterranean species of blister beetles, *Lydus trimaculatus* and *Mylabris variabilis* (Coleoptera: Meloidae)

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Abstract

Cantharidin (CA), a toxic terpene produced by blister beetles (Coleoptera: Meloidae), attracts the interest of many researchers for its renowned medical properties. The CA content in blister beetles has been mainly quantified in some Oriental species, due to their use in traditional Chinese medicine, or in few other species of toxicological importance. As CA quantification has been largely ignored in many other members of this family, we aimed at estimating CA content in natural populations of two of the most abundant species in central Italy, *Mylabris variabilis* and *Lydus trimaculatus*. Nearly 100 individuals for each species were collected in the field, identified and reared in fauna-boxes until CA collection. Available protocols were optimized for CA extraction from both dried-body tissues and exuded hemolymph collected from leg joints using capillary tubes. The CA content was quantified in a gas chromatography system coupled to a mass spectrometer using a calibration curve with diethyl-ester of norcantharidin as the internal standard. We observed: (i) high variability in CA content among specimens, which was positively related to individual dry-weight; and (ii) slightly larger (though non-significant) amount of CA in males than in females for both species. Our data are consistent with the available published reports about CA content, transfer and distribution in blister beetles and suggest a conserved biological role of this terpene in Meloidae.

Key words: cantharidin, defensive terpene, GC-MS, hemolymph, toxicity.

INTRODUCTION

Meloidae, or "blister beetles", is a widespread family of Coleoptera Tenebrionoidea which includes almost 3,000 species (Bologna 1991; Bologna *et al.* 2008, 2010), all characterized by the ability to produce and store cantharidin (CA), a toxic terpene. When disturbed, blister beetles usually enter thanatosis and secrete CA as yellow oily droplets of hemolymph from leg joints ("reflex bleeding"). Hence, CA is supposed to have a primary defensive role, but also to be involved in reproduction. In fact, males of meloid beetles usually produce more CA than females (Capinera *et al.* 1985; Blodgett *et al.* 1991; Carrel *et al.* 1993; Nikbakhtzadeh & Tirgari 2002; Mebs *et al.* 2009; Bravo *et al.* 2017) and transfer large quantities of this terpene to females during copulation (Selander 1964; Bologna 1991; Carrel *et al.* 1993; Dettner 1997; Nikbakhtzadeh 2004; Nikbakhtzadeh *et al.* 2012). Females are supposed to use the transferred CA for covering eggs to protect them from potential predators (Sierra *et al.* 1976; Carrel *et al.* 1993; Eisner *et al.* 2002).

Cantharidin properties have been independently discovered by distinct ancient cultures and used for many purposes in popular pharmacology for more than two millennia (Bologna 1991). Traditionally considered as a sexual stimulant (Beauregard 1890; Wang 1989; Moed *et al.* 2001), CA is nowadays used for the topical

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