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Four types of malformations in a population of *Bufo boulengeri* (Amphibia, Anura, Bufonidae) from the Jbilet Mountains (Marrakech, Morocco)

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Amphibians are considered as excellent "bio indicators" of environmental health (Schuytema and Nebeker, 1999; Tejedo, 2003; García-Muñoz et al., 2010). Although malformed amphibians have been recorded for nearly 300 years (e.g. Ouellet, 2000), the current increase of this phenomenon is remarkable. While abnormalities arising from mutation, developmental errors and trauma are to be expected in any amphibian population, they typically appear in low proportions (< 5%), and most often involve only missing digits or limb parts (Blaustein and Johnson, 2003).

Different causes have been proposed to explain this increase of the malformations rate in amphibian populations. Recent evidence points to the infection by a parasitic flatworm (*Ribeiroia ondatrae*), as the widespread cause for limb malformations in amphibians, particularly supernumerary limbs (e.g. Sessions and Ruth, 1990; Johnson et al., 1999). Throughout evolutionary history, UV radiation has been an ubiquitous stressor on living organisms (Cockell, 2001), and some research has shown that UV-B radiation can induce a variety

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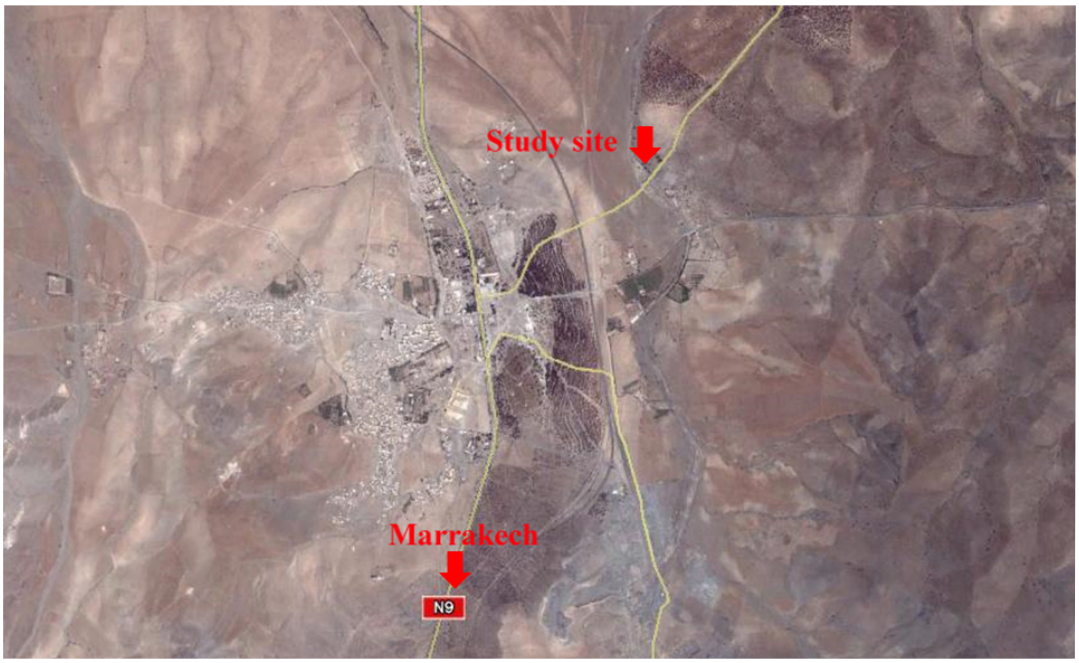

Figure 1. Studied pond situated in the Jbilet Mountains, near Marrakech.

Figure 2. Recent metamorph of *B. boulengeri* with a supernumerary hind limbs deformity.

of physical abnormalities in amphibians (Ankley et al., 2002). Chemical contaminants are perhaps the most alarming causative factor for the increase of developmental problems (Blaustein and Johnson, 2003), and numerous laboratory studies have shown that many different contaminants can kill or cause deformations in amphibians (Sparling, Linder and Bishop, 2000; Mahapatra, Mohanty-Hejmadi and Chainy, 2001; Piha et al., 2006).

In this study a pond situated in the Jbilet Mountains, near Marrakech (Morocco; Latitude: 31.907702; Longitude: -7.933901; 480 m a.s.l.; Figure 1) was examined in search of amphibians in May 2010. Two anuran species were found, both widely distributed in Morocco and co-occurring in this area (Bons and Geniez, 1996): *Bufo boulengeri* (Lataste, 1879) (according to Stöck et al., 2006, 2008) and *Amietophrynus mauritanicus* (following Van Bocxlaer et al., 2009).

A total of 30 newly metamorphosed specimens of both species (*B. boulengeri*, N=12; *A. mauritanicus*, N=18) randomly selected were analysed. Metamorph characterization was restricted to external examination of the individuals. Thus, other abnormalities could have remained undetected without the use of radiography and histopathology.

Two different malformed individuals, both corresponding to *B. boulengeri*, were detected holding a total of four different types of malformations. The first individual (Figure 2), presented a supernumerary hind limbs deformity, and the second one (Figure 3),

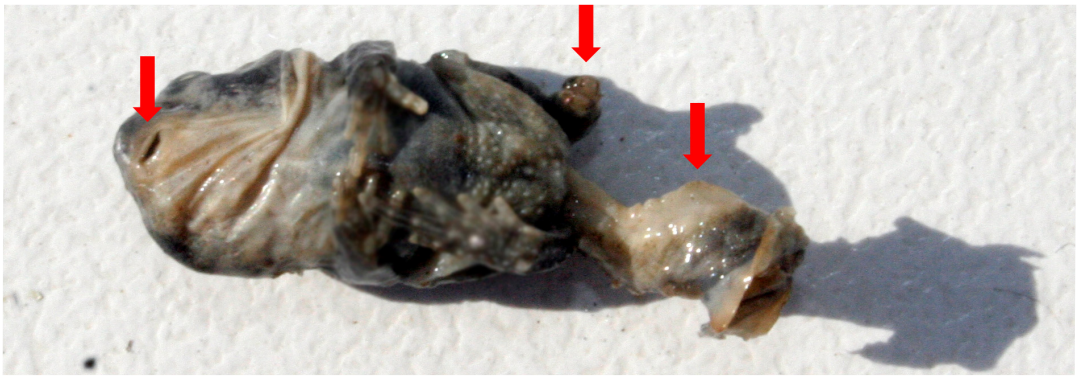

Figure 3. Recent metamorph of *B. boulengeri* with three types of malformations: oral deformities, him limb deformities and incomplete tail reabsorption.

presented the general aspect of a metamorph, but with the mouth structure of the larval stage, no toe in the left hind limb, and incomplete tail reabsorption.

Some previous studies have described malformations in the European members of the *B. viridis* species group associated to environmental disturbance (Henle, 1981; Borkin and Pikulik, 1986; Flyaks and Borkin, 2004). However, to our knowledge this is the first report in *B. boulengeri*. Although we did not investigate the cause of these malformations, the easiness such malformed metamorphs were detected and the unusually high malformation rate (16.6%) in this population of *B. boulengeri* point to an environmental disturbance. In fact, the use of wetland as a landfill, the high livestock pressure (authors pers. obs.), and discharge of wastewater into the wetland from nearby residential areas, represent potential factors for the observed malformations.

High rates of abnormalities in amphibians have been associated with declining populations (Cohen, 2001). Namely, massive mortality of eggs, larvae, and adults, often occur together with deformed individuals only rarely surviving to adulthood (Johnson et al., 2001). The potentiality of malformations as signals of ecosystem disruption, and the secondary effects that this potential disruption might have on other organisms sharing the same ecosystems, is a matter for future ecotoxicologycal research. Further work at this site (and others undergoing similar problems) should include monitoring, and wetland conservation plans in order to reduce the direct impact of human activities (García-Muñoz et al., 2010).

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