INTRODUCTION

Honey bees are wild or semi-domesticated insects that live in colonies with complex division of labor (Winston, 1987). Bee colonies consist of one egg-laying queen, the female worker caste, which includes nurse bees and foragers, and the male drones. In contrast to queens and drones, the life-histories of workers can be very plastic during adulthood - individuals change from one social care task to another (Winston and Fergusson, 1985). In summer, workers typically progress from nurse bees with in-hive tasks to foragers. In autumn, when brood production decreases, bees develop into so-called winter bees (Maurizio 1950). Besides physiological specialization enabling bees to perform different tasks, the three major worker types (nurse, forager and winter bee) differ markedly in life-span. Foragers, one type of worker bee, typically die within two weeks and are the shortest-lived individuals, whereas bees continuing nursing can have life-spans longer than 50 days. The longest-lived workers, winter bees, survive from late summer to next spring. Only the queen lives longer, surviving 2 to 3 years, and in some cases up to 5 years (Dukas 2008, Münch et al. 2013).

The transition of nurse bees to foragers is hormonally regulated by juvenile hormone and vitellogenin. During the life course of workers, vitellogenin levels in the hemolymph and fat body drop, and these reduced concentrations influence several aspects of the life history of bees (Münch and Amdam, 2010). Levels of vitellogenin in hemolymph and fat body are highest in the long-living winter bees (up to 60-90 μg/μL hemolymph), and lowest in short-living foragers (0-5 μg/μL hemolymph) (Seehuus et al. 2006).

The phospholipoglycoprotein (Wheeler and Kawooya 1990) vitellogenin is a female-specific egg-yolk precursor (Spieth et al. 1991), synthesized by most oviparous animals including insects for transfer into oocytes, where it serves as nutrition for embryos. Besides the full length 180 kDa protein, vitellogenin was reported to occur as a 150 kDa fragment (Seehuus et al. 2007) and a 40 kDa fragment (Havukainen et al. 2010) in honey bees. Vitellogenin

is also found in the non-reproductive worker castes of bees. It is produced in the fat body (a tissue functionally homologous to the white adipose tissue and liver of vertebrates) in the abdomen (Snodgrass, 1956). Vitellogenin is generally released from the fat body, circulates in the hemolymph and is taken up by the ovaries through receptor-mediated endocytosis (Amdam et al., 2003; Guidugli et al. 2005). Honey bee workers typically do not import vitellogenin to the ovaries, but vitellogenin can be taken up for example by the hypopharyngeal glands in the head of nurse bees that synthesize food jelly for young larvae, other workers and the queen (Amdam et al., 2003). Vitellogenin also shields cells from oxidative damage and protects both workers and queens from oxidative stress (Seehuus et al. 2006; Corona et al. 2007; Havukainen et al. 2013).

Several factors such as pathogens and plant protection products (PPPs, pesticides) can alter vitellogenin levels in honey bees. Infection of young nurse bees with *Nosema ceranae* decreased the expression of vitellogenin transcript (Antúnez et al. 2009) whereas infection of honey bee larvae caused increased vitellogenin titers in young adults (BenVau and Nieh 2017). Some PPPs, including neonicotinoids, the organophosphate chlorpyriphos and the pyrethroid cypermethrin led to increased levels of the *vitellogenin* transcript in the brain of mixed-aged honey bee workers exposed in the period between beginning of May and end of June (Christen et al. 2016, Christen and Fent 2017). In contrast, the organophosphate malathion and chlorantraniliprole decreased the *vitellogenin* transcript (Christen et al. 2017).

The aim of our present study is to assess whether the induction of vitellogenin mRNA upon exposure to the neonicotinoid clothianidin is paralleled on the protein level. First, we generated and characterised an *Apis mellifera* vitellogenin antibody and evaluated its presence in different tissues. Second, we analysed vitellogenin protein levels in different tissues upon exposure to the neonicotinoid clothianidin and compared them with mRNA levels. We detected tissue-specific expression patterns of vitellogenin proteins. Our study demonstrates that previously reported alterations in vitellogenin mRNA are accompanied by changes in protein