1 INTRODUCTION

Aphids (Homoptera: Aphidoidea) are economically one of the most important agricultural pests. In the last years, some of aphids such as *Aphis fabae* SCOPOLI, *Aphis gossypii* GLOVER, *Acyrthosiphon pisum* (HARRIS), *Myzus persicae* (SULZER) and *Aphis pomi* (DE GEER) have been very important pest species of many greenhouse and field plants in Europe. They are phytophage cosmopolitan and polyphagous species that sucking of plant phloem sap and transmission of plant viruses (POWER 1987, RASSABY et al. 2004). Moreover, aphids easily develop insecticide resistance (HERRON et al. 2001, KIFT et al. 2004). Nowadays, environmental hazard due to regular and rather intensive chemical insecticide spraying is a growing concern. Thus, biological control of aphids using natural enemies, seems to be a successfully control allowing the amount of insecticides to be reduced, is needed to create sustainable agriculture development (PUTRA and YASUDA 2006).

Today, there are many different biological agents, which are suitable for a biological control of aphids. In the last years, several studies have conducted on the predatory flower bugs *Orius* spp. (Heteroptera: Anthocoridae). The *Orius* spp. have many characteristics of ideal biological control agents, i.e., high searching efficiency, an ability to increase more rapidly when prey is abundant, a density-dependent decrease in fecundity resulting from interference and the ability to aggregate in regions of high prey density (HODGSON and AVELING 1988). They are generalist predators attacking a variety of small arthropods such as aphids, aleyrodids, young lepidopterous larvae, thrips and mites (BARBER 1936, PÉRICART 1972). Due to their effectiveness in a number of agricultural ecosystems, several species have received considerable attention as biological control agents (COCUZZA et al. 1997a). Some *Orius* bugs are already used as biological control agents against *Frankliniella occidentalis* (PERGANDE), on vegetable and ornamental crops under greenhouse conditions in Europe (VAN LENTEREN et al. 1997).

Orius similis ZHENG which originated from China is a common bug species that preys on different aphid species, *Frankliniella formosae* MOULTON, *Tetranychus cinnabarinus* (BOISDUVAL), eggs or hatched larvae of *Pectinophora gossypiella* (SAUNDERS), *Helicoverpa armigera* (HÜBNER) and *Anomis flava* (FABRICIUS) as well as on other Lepidoptera pest and it can also feed upon plant pollen (ZONG et al. 1987, ZHANG et al. 1994, ZHOU and LEI 2002). This predatory bug has been recorded as an effective predator of eggs of *P. gossypiella* in China (ZONG et al. 1987, ZHANG et al. 1994). Also, it was used to control the cotton aphid, *A.*

gossypii in cotton field (ZHOU and LEI 2002). Recently, *O. similis* seems to be a promising predator of different aphid species (AHMADI et al. 2008a, b, SENGONCA et al. 2008). However, such sufficient knowledge of *O. similis* is still lacking in the literature.

Studies of the biological and ecological characteristics of a natural enemy must be undertaken prior to its mass-production and release as a biological control agent. The ability of a natural enemy not only in terms of its predatory potential but also in its adaptability to different environmental conditions are the essential prerequisites for the successful utilization in biological control programs. Also, its preference for different prey species as well as cannibalism and intraguild predation with other natural enemies should be determined. In addition, studying of oviposition and egg-laying behavior strategies of a natural enemy is of a great value leads to a better understanding of its biological and ecological characteristics, which determines the fitness of its offspring and growth rate in the population (DANHO and HAUBRUGE 2003). Moreover, in order to integrate the use of pesticides with natural enemies that are used in IPM programs, the side effect of pesticides to the beneficial arthropods must be established. Finally, releasing of a natural enemy under more natural conditions is necessary to evaluate its efficiency to control a given pest species.

Therefore, the present work aimed to study the biology and prey consumption of the polyphagous predatory bug *O. similis* at 18±1°C, 25±1°C and 30±1°C constant temperatures with different aphid species as prey in the laboratory. Further experiments were devoted to record the prey consumption preferences, longevity of the predator with different nutritional sources as well as the egg-laying preferences of the predator. Also, the cannibalism by the predator and intraguild predation between *O. similis* and *Dicyphus tamaninii* WAGNER (Het., Miridae) was stated at different population densities of aphid species. Additionally, the experiments were conducted to investigate the toxicity of selected pesticides on the predator as well as effect of indoxacarb and pirimicarb on its biology and prey consumption. Finally, greenhouse experiments were carried out in order to confirm the efficiency of *O. similis* for the biological control of *A. gossypii* on protected broad bean plants.

2 MATERIAL AND METHODS

2.1 Laboratory experiments

2.1.1 Rearing of insects

2.1.1.1 Rearing of aphid species as prey

The stock cultures of *A. fabae* and *A. pisum* on broad bean, *A. gossypii* on cotton, *Schizaphis graminum* (RONDANI) and *Sitobion avenae* (FABRICUS) on wheat and *M. persicae* on cabbage plants were established with individuals obtained from stock cultures available at the INRES-Phytomedicine, University of Bonn, Germany. The stock culture was kept in a climatically controlled chamber at 25±1°C temperature, relative humidity of 60±10% and 16 hours of artificial light at an intensity of about 4000 lux. The host plants were usually planted in treys (60×40 cm) and had been replaced with new ones fortnightly or whenever more aphid species was needed for the experiments. The aphid species on the leaves of the old plants were used to infest the new plants as well as to feed the predator. The apple aphid, *A. pomi* and the walnut aphid, *Chromaphis juglandicola* (KALTENBACH) used in the experiments were originally obtained from the experimental teaching garden of INRES-Phytomedicine, University of Bonn, Germany.

In order to obtain the 1-2- and 4-5-day-old as well as adult of different aphid species required, up to ten adult virginoparae aphid species females were placed on the freshly excised host plant leaf discs (2.5 cm diameter) were placed in the round plastic Petri dishes (3.5 cm diameter) for nymph laying. The round plastic Petri dishes were filled with 0.5 cm-thick-layer of 0.7% agar gel, and with a meshed hole in the lid to allow air exchange (Fig. 1).

Fig. 1: Round plastic Petri dish using for the obtaining of the uniformly aged different aphid species and the experiments for *Orius similis*

The agar served as a support and kept the leaf discs green and fresh. After 24 hours, the adults were gently transferred into new similarly prepared round plastic Petri dishes by using a camelhair brush. The obtained nymphs were reared further until reaching the age desired for the experiments. The nymphs were usually kept in an incubator at 25±1°C temperature, relative humidity of 60+10% and a photoperiod of 16:8h (L:D).

2.1.1.2 Rearing of *Orius similis*

A colony culture of *O. similis* was initiated from a few individuals obtained courtesy from Fujian Academy of Agricultural Sciences (FAAS), in Fuzhou, PR China. They were reared on freshly excised broad bean leaf discs 2.5 cm in diameter, which were placed upside down onto, the round plastic Petri dishes 3.5 cm in diameter as mentioned in capital 2.1.1.1. Broad bean leaves were infested with more than 20 nymphs of *A. pisum* as prey. The round plastic Petri dishes were held in a controlled climate chamber at 25±1°C temperature, 60±10% relative humidity and a photoperiod of 16:8h (L:D) with an artificial light intensity of about 4.000 lux. Adult mated females were transferred separately to round plastic Petri dishes for oviposition. After 48h, the adult females were moved to other round plastic Petri dishes and the laid eggs were incubated until egg hatching. These leaves and N_1 nymphs were placed into the Plexiglas cages (15×7.5×4.5 cm), with three mesh-covered holes in the lid (Fig. 2), to start the pre-imaginal rearing.

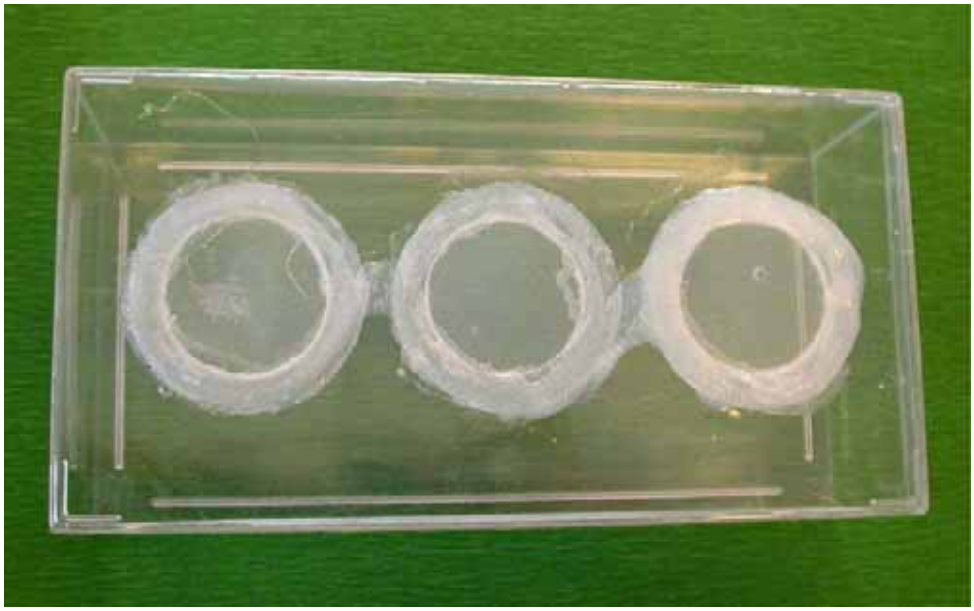

Fig. 2: Plexiglas cage using for the pre-imaginal rearing of *Orius similis*

The Plexiglas cages were kept in an incubator under the same condition mentioned above until *O. similis* reached the adult stage. The leaves and stems of broad bean plants infested with *A. pisum* were served as substrate for rearing the predator. To maintain adequate prey supply continuously, the substrates infested with the prey species were frequently replaced inside the cages. All the populations were reared in the laboratory for five to seven generation before the start of the experiment.

2.1.1.3 Rearing of other insects and mites

2.1.1.3.1 Prey species

To determine prey consumption preferences of *O. similis*, several economically important arthropod pests of greenhouse and field crops were reared and tested as possible prey species of the polyphagous predatory bug. These were in addition to different aphid species, the two-spotted spider mite, *Tetranychus urticae* KOCH (Acari, Tetranychidae), the western flower thrips, *F. occidentalis* and the cotton whitefly, *Bemisia tabaci* (GENN.) (Hom., Aleyrodidae).

All the stock cultures of the prey species were established with individuals obtained from stock cultures available at the INRES-Phytomedicine, University of Bonn, Germany. The stock cultures of *T. urticae* and *F. occidentalis* were maintained on bean plants (*Vulgaris phaseolus* L., cv. "Marona"), while the cotton whitefly, *B. tabaci* was reared on cotton plants. Heavily infested bean and cotton plants were usually replaced with fresh ones. The rearing of the prey species took place in the climatically controlled chambers at 25±1°C temperature, 60±10% RH and 16:8h (L:D) photoperiod.

The desired stages of the prey species for the experiments were identified under a binocular microscope in the laboratory on leaves obtained from the different host plants in the different stock cultures and then gently collected by using a camelhair brush.

2.1.1.3.2 *Dicyphus tamaninii*

For the experiment on the intraguild predation between *O. similis* and other predator as well as the experiments on the oviposition of *O. similis* in the presence of other natural enemy, the predatory bug, *D. tamaninii* was selected. Individuals of *D. tamaninii* had been obtained from stock cultures available at the INRES-Phytomedicine, University of Bonn, Germany. The predator was maintained on broad bean plants infested with *A. pisum* as prey in a cage

(60×60×40 cm) sealed with gauze from four sides in order to allow aeration. The stock cultures were kept in climatic control chambers as mentioned per aphid species in capital 2.1.1.1. For obtaining of individuals in the desired age, the hatched nymphs were gently collected by using an aspirator and transferred on broad bean leaf discs infested with more than 30 nymphs of *A. pisum* as prey placed in the round plastic Petri dishes (3.5 cm diameter) as described above in capital 2.1.1.2. The nymphs were reared further until reaching the age desired for the experiments.

2.1.2 Procedures of the experiments

2.1.2.1 Biology and prey consumption of *Orius similis*

2.1.2.1.1 Biology of *Orius similis*

The biology of *O. similis* was comprehensively investigated with different aphid species as prey in the laboratory at 25±1°C temperature, relative humidity of 60±10% and a photoperiod of 16:8h (L:D). For all the experiments on embryonic and nymphal development, mortality, longevity as well as fecundity, round plastic Petri dishes 3.5 cm in diameter as seen in figure 1 were used. The round plastic Petri dishes contained broad bean leaves as common host plant of *A. fabae*, *A. gossypii*, *A. pisum* and *M. persicae* or apple leaves as host plant of *A. pomi*. All populations of predator were reared in the laboratory for five to seven generations before the start of the experiments.

2.1.2.1.1.1 Embryonic and nymphal development as well as mortality

In order to determine the duration of embryonic development, 20 adult mated females of *O. similis* (3, 10 and 17 days after starting oviposition) were confined singly on the freshly excised host plant leaf discs (2.5 cm diameter) which were placed upside down onto the agar gel layer for 24 hours for egg laying. The females were offered 20 individuals of 1-2-day-old different aphid species as prey. After egg laying, both *O. similis* females and aphid species were removed and the leaf discs with newly laid eggs were incubated and checked daily until the hatching of N_1 nymphs. The duration of embryonic development and percentage mortality of eggs were recorded after hatching the nymphs from the eggs. The newly hatched 1^{st} instars were singly transferred with a camel hair brush into other similar round plastic Petri dishes containing freshly excised host plant leaves infested with 20 individuals of 1-2 day-old different aphid species as prey. The round plastic Petri dishes were checked daily for

moulting and the mortality of *O. similis* nymphs. The nymphal instars were transferred to new round plastic Petri dishes every day until the adult stage. A minimum of 30 replicates was set up with each prey species.

2.1.2.1.1.2 Longevity

For recording the longevity of *O. similis* unmated females and males, not less than 50 individuals of the last nymphal instars were taken and placed singly in the round plastic Petri dishes as described above, newly emerged adult females and males were daily provided with an excess number (20 individuals) of 1-2-days-old different aphis species nymphs as prey until they died. In order to determine the longevity of mated females, the newly emerged females and males were transferred within a period of two days to new round plastic Petri dishes for mating, mated females were transferred to other round plastic Petri dishes and reared until death. At least 12 adult mated females were used for each prey species.

2.1.2.1.1.3 Fecundity

2.1.2.1.1.3.1 Oviposition period

During the investigation of the longevity, the first and last day of egg laying of mated females were also recorded for evaluating the pre-oviposition, oviposition and post-oviposition periods.

2.1.2.1.1.3.2 Daily and total fecundity by feeding different aphid species

In order to establish the fecundity of *O. similis* females during their ovipostion period, the numbers of laid eggs were recorded and the round plastic Petri dishes changed daily. The daily and total fecundity and the mean daily oviposition were determined. The mean daily oviposition of each replication was calculated as mean total fecundity in oviposition period.

2.1.2.1.1.3.3 Oviposition at different environmental condition

Fecundity of natural enemies is an important component of fitness. It varies with biotic and abiotic factors, which can affect their use in biological control programs. Thus, investigation of oviposition of a natural enemy at different environmental condition is of a great value leads to a better understanding of its ecological characteristics and explaining the oviposition strategy, which determines the fitness of its offspring and growth rate in the population.