GROWTH AND IONIC COMPOSITION OF THE ANTENNAE OF VARIEGATED GRASSHOPPER, *ZONOCERUS VARIEGATUS* (L., 1758) (ORTHOPTERA: PYRGOMORPHIDAE).

K. O. Ademolu*, A. A. Balogun and A. B. Idowu

* Biological Sciences Department, University of Agriculture, Abeokuta P.M.B 2240, Abeokuta, Ogun State, NIGERIA. E-mail: kennyademolu@yahoo.com

[**Ademolu, K. O., Balogun, A. A. & Idowu, A. B.** 2011. Growth and ionic composition of the antennae of variegated grasshopper, *Zonocerus variegatus* (L., 1758) (Orthoptera: Pyrgomorphidae). Munis Entomology & Zoology, 6 (2): 920-924]

ABSTRACT: This study examines the growth pattern and the ionic composition (Na^+, K^+, Ca^{2+}, Mg^{2+}, Zn^{2+}, Fe^{2+}, Po_4^{2-},Cl^-) of the antennae of *Zonocerus variegatus* during its post embryonic development. The antennal length (cm) and number of antennal segments increased steadily during development with the adult stage having significantly ($P<0.05$) higher values than the lower instars. The growth gradient between the 6th instar and the adult stage was greater than that within the lower instars. Also, the growth rate of the body length (0.415x) was faster than the antennal length (0.215x). The 1st and 2nd instar stages of development had significantly higher ionic concentrations than the later instars and the adult stage. The relationship between these results and the behavior of the *Z. variegatus* is discussed.

KEY WORDS: *Z. variegatus*, antennae, growth rate, ionic composition.

The antennae are the most important sensory organs for insects, bearing not only the sensilla of olfaction, but also of those of taste, mechano, hygro, thermo perception and sometimes sensors of CO_2 (Hansson, 1999).

For many insects, antenna is of utmost importance not only in search for food for themselves or their offspring, but for intraspecific communications as well (Hansson, 1995; Zacharuk & Shields, 1991).

In *Zonocerus variegatus* and other acrids, the size of antennae and antennal lobe increased with each moult from first instar to the adult stage (Youdeowei, 1974) with special sensillum types such as pheromones receptors finally arising in the adult.

The attraction of *Z. variegatus* to siam weed *Chromolaena odorata* (L.) leaves and inflorescences in the field had been attributed to olfactory clues and not visual (Modder, 1984) indicating that the antenna is used in the response.

The life cycle of *Z. variegatus* is built around six nymphal stages and adult stage (Toye, 1982; Yondeowei, 1974) and the nymphal development is usually completed in 3-5 months, while the adults lives up to 15 weeks (Muse, 2003). The behavior and food preferences of each nymphal stage differs: while the 1st-3rd instars have gregarious habit, aggregating or crowding around *C.odorata* and prefer weeds like *C. odorata, Ageratum conyzondes, Aspillia lacifolia, Tridax procubens*, later instars on the other hand, are dispersed and prefer cassava (*Manihot esculenta*) leaves (Chapman, 1986; Tamu, 1990). These differences in the life cycle of grasshoppers are coupled to differences in morphological and physiology features of the sensory organs (Anton et al., 1996).

In the recent times, electrophysiology and electroantennogram (EAG) had been used extensively in insect pest control and it gives a good knowledge of the insect and of host-plant interaction which could be used in producing an effective trap-bait system.

However, there is scarce information on the physiology of the antennae of *Z. variegatus* in literature the little present are on its morphometrics and descriptions (Youdeowei, 1974 and Chapman et al., 1977). The focus of this study is to examine the growth and ionic composition of the antennae of *Z. variegates*.

MATERIALS AND METHODS

Newly hatched first instar nymphs of Z. variegatus were collected from uncultivated farm land near Students Union Building of University of Agriculture, Abeokuta, Nigeria. They were reared in wire mesh cages (30x30x45cm) situated at the insectary of the Department of Biological Sciences of the University. They were maintained till they reached adult stage on fresh leaves *C. odorata* and *M. esculenta*. The identification of each of the six nymphal stages was done based on the description of Chapman et al., (1977) and Youdeowei (1974). After each moult, evidenced by the exuviae, antennectomy was done on each instar stage. This was done with fine scissors and it involved the removal of the whole flagella and pedicel segments of both sides.

Measurements

The antennal length and number of segments each of the antennae of each nymphal stage was measured using calibrated microscope. The body weight of each stage of development was taken using sensitive electronic balance (Mettler PM-11 UK). 30 replicates of each nymphal stage were used.

Ionic Analysis

The ionic composition of the antennal was determined by method of A.O.A.C (1990). 0.5g of the antennal sample of each instar stage was oven dried and well ground samples were weighed into 250ml flask for wet ashing by the addition of 25ml of HNO_3, 4ml of HCl and 2ml of H_2SO_4. This mixture was then heated for 5 minutes and diluted with distilled water to 100ml. Magnesium d(Mg^{2+}), Zinc (Zn^{2+}) calcium (Ca^{2+}) and iron (Fe^{+}) were determined by using Atomic Absorption spectrophotometry (AAS) (Pye unican UK model SP9). Sodium (Na^{+}) and potassium (K^{+}) were also determined through flame photometer (Corning, UK model 405) while phosphate (Po_4^{2-}) and chloride (Cl^{-}) were analyzed by colorimetric method using spectronic 20 (Gallenkamp, UK). All these analyses were done in triplicates.

Statistical Analysis

The data collected were analyzed by one-way analysis of variance (ANOVA) and significantly different means were separated using Student-Newman-Keuls (SNK) test. Regression analysis was used to determine the growth rate of the body parts and the relationship among the measured parameters.

RESULTS

There was a gradual increase in the length and number of segments on the antennae as the insect moult from 1st instar to the adult stage, the antennal length of the 1st instar was 0.19 cm while the adult was 1.75cm. Similar trend was recorded for the body length and body weight (Table 1). The adult stage had significantly higher ($P<0.05$) antennal length than other early stages of development, while the antennal segments of 5th instar to adult stage were significantly more than those of 1st-3rd instars.

The growth gradient or slope between the 6th instar and adult stage was greater than that within other lower stages of development (Figure 1). Similarly, the growth rate of the antennal length (0.2154x) was slower than the growth rate of body length (0.415x). Furthermore, regression analysis reveals a strong and positive relationship between the antennal length and body weight (+0.96) (Figure 2).

The ionic composition of the antennae of *Z. variegatus* during post embryonic development is shown in table 2. There were significant differences ($P<0.05$) in the concentrations of the ions across the developmental stages. The concentrations of Na^+, K^+, Mg^{2+}, Zn^{2+}, Ca^{2+} were significantly higher in early instars (1st-2nd) than the latter instars (5th-adult). However, no significant difference ($P>0.05$) was observed in the concentrations of the anions (Po_4^{2-} and Cl^-) across the developmental stages. Of the eight (8) ions detected in the antennal of *Z. variegatus* during development CL^- followed by K^+ recorded the highest concentrations. Furthermore, the total concentration of ions in the 1st and 2nd instars were 88..13 mg/mol and 87.64 mg/mol respectively while the 6th and adult stage antennae had total concentration of 51.83mg/mol and 69.56mg/mol respectively (Table 2).

The total concentration of ions in the antenna of each developmental stage is shown in Figure 3. The 1st and 2nd instar stages recorded higher total concentrations of ions than later instars. A drop in total concentration was observed at the 3rd instar stage which rose up again at the 4th instar stage, but later dropped during 5th and 6th stages of development. The adult stage however witnessed a rise in the total concentration but not as high as that of the 1st and 2nd instar stages of development.

DISCUSSION

The length of the antennae and number of segments on them progressively increased during post embryonic development. Chapman *et al.,* (1977) made similar observation on *Z. variegatus* fed different food plants in Ibadan, Nigeria. This is also in agreement with the results of other electrophysiologists (Anton et al., 2002; Anton et al., 1996 and Hansson et al., 1996) that antennal lobe of *Schistocerca gregaria* increased in size during development due to increased number and size of glomeruli.

The size of the antennae might provide explanation for the differences in the behavior of *Z. variegatus* during development as the short length of the antennae of the lower instar (1st-3rd) might not allow them to perceive or detect very well odor far off from them thus resulting in the gregarious or crowding habit. On the other hand, the long antennal length of the adult stage allows for easy perception of both near and far odor making them to be dispersed widely in the field. Changes in the peripheral olfactory input between developmental stages had been reported to cause a behavioral switch in *S. gregaria* (Ochieng' et al., 1998). During post embryonic development of Cricket *Gryllus bimaculatus* both the quantity and quality of sensory inputs periodically change at the time of ecdysis. The longer cercal filiform hairs are more sensitive to air current than short ones in the adult stage (Kanou et al., 1988). In a related experiment, Wasserman and Itagaki (2003) observed that maxillary palps of Flesh fly, *Neobelleiria bullata* (Diptera: Sarcophagidae) were less sensitive to plant oriented odorants than the antennae because of the anatomical position relative to the source of the odour.

In many hemimetabolans, additional antennal flagellomeres and sensilum are formed with pheromones receptors finally arising in the adult stage (Hansson, 1999). The present study correlated this fact as the growth slope between the 6th conster and adult stage was greater than that of the lower instars. This is consistent with the increasing number of reception neurons during post embryonic development. This phenomenon might perhaps be due to the reproductive activities that in pronounced and unique in the adult stage of *Z. variegatus* which specifically requires the use of antennae for courtship (mate location and identification) and mating behaviors in addition to food selection peculiar to all instars.This parallels the observation of Ochieng' et al. (1998) that the number of olfactory sensillum reached a significantly higher number at the last stage of development in gregarious adult locust, *S. gregaria*. Similarly, the enzymatic activities of the adult *Z.variegatus* femoral muscles were significantly ($P<0.05$) higher than the early instar stages (Ademolu et al., 2009).

Antennal length forms a good indication or reflection of the body weight of *Z. variegatus* as there existed a strong and positive relationship between the two body measurements. Idowu (1995) likewise reported that a strong relationship existed between the body size and the repellent gland size of *Z. variegatus*. Thus, in laboratory experiments, antennal length could serve as an estimate of body weight if weighing balances are not available.

The ionic concentrations of antennae of the lower instar (1st-2nd) were significantly higher than the later stages of development. Similarly, high concentrations of K^+, Na^+ and Cl^- in the antennae of early instars are noteworthy. Na^+ and K^+ are known to play vital roles in transmission of impulses in the cells (Ademolu et al., 2007). The higher concentrations of these ions in the lower instars might probably be accountable for the gregarious and quiescent nature at these early stages of development, because high concentrations of these ions leads to decrease in resting potential of the cells and thus reduce their sensory function (Chapman, 1990). Ignell et al., (1998) similarly observed that projection neurons (PNS) of the adult gregarious locust, *S. gregaria* responded significantly more frequently to phenylacetonitril (PAN) than PNS of lower gregarious instars.

High concentrations of K^+ and Cl^- were observed in the antennae of *Z. variegatus* in their study. This is not unexpected as phytophagous insects like *Z. variegatus* commonly have high concentrations of K^+ and Cl^- in their tissues due to the plant origin of their foods (Chapman, 1990).

CONCLUSION

We have been able to show both morphologically and physiologically that the later instars and adult stage of *Z. variegatus* will be more sensitive than the lower instars. We therefore opine that traps baited with attractive odor will best work for later instars and adult stage that have higher response capability or excitability than the lower instars. Hence, use of olfactory pheromone will do better in later stages of development than lower instars where other IPM techniques might be applied.

LITERATURE CITED

Ademolu, K. O., Idowu, A. B. & Olatunde, G. O. 2009. Morphometrics and enzymes activities in the femoral muscles of variegated grasshopper *Zonocerus variegatus* (Orthoptera: Pgrgomorphidae) during post embryonic development. International Journal of Tropical Insect Science, 29: 53-56.

Ademolu, K. O., Idowu, A. B. & Amusan, A. A. A. 2007. Chemical analysis of tissues of *Zonocerus variegatus* (L.) (Orthoptera: Pyrgomorphidae) during post embryonic development in Abeokuta, south-western, Nigeria. Nigerian Journal of Entomology, 24: 24-34.

Anton, S., Ignell, R. & Hansson, B. S. 2002. Developmental changes in the structure and function of the central olfactory system in gregarious and solitary desert locusts. Microscopy Research, and Techniques, 56: 281-291.

Anton, S. & Hansson, B. S. 1996. Antennal lobe interneurons in the desert locust *Schistocerca gregaria* (Forskal). Processing of Aggregation pheromones in Adult male and female. Journal of comparative neurology, 370: 85-96.

A. O. A. C. 1990. Association of official analytical chemist (ed. W. Hormitz 13th ed. Washington. 1142 pp.

Chapman, R. F. 1990. The insert: structure and function. The English language. Book society and Hodder and Stoughton, Great Britain. 968 pp.

Chapman, R. F. 1986. Bionomics of the variegated grasshopper (*Zonocerus variegatus)* in West and Central Africa. Annual Review of Entomology, 31: 479-505.

Chapman, R. F., Cook, A. G., Mitchell, G. A. & Page, W. W. 1977. Description and morphometrics of nymphs of *Zonocerus variegatus (*Orthoptera: Acridoidea). Bulletin of Entomology Research, 67: 427-437.

Hansson, B. S. 1995. Olfaction in Lepidoptera. Experiential, 51: 1003-1022.

Hansson, B. S., Ochieng', S. A., Grosmaitre, Y., Anton, S. & Njagi, P. G. N. 1996. Physiological responses and central nervous projections of antennal olfactory receptor neurons in the adult desert locust *Schistocerca gregaria* (Orthoptera: Acrididae). Journal of comparative physiology, A 179: 157-167.

Hansson, B. S. 1999. Insect olfaction. Springer-verlag Berlin Heidelberg, Germany. Pp. 16-24.

Idowu, A. B. 1995. Structure of the repellent gland of *Zonocerus variegatus*. Journal of African Zoology, 107: 247-252.

Ignell, R., Anton, S. & Hansson, B. S. 1998. Central Nervous processing of behavioural relevant odours in solitary and gregarious fifth instar locust *Schistocerca gregaria*. Journal of comparative physiology, A. 183: 453-465.

Kanou, M., Osawa, T. & Shimozawa, T. 1988. Ecdysial growth of the filiform hairs and sensitivity of the cercal sensory system of the cricket *Gryllus bimaculatus*. Journal of comparative physiology, A. 162: 573-579.

Modder, W. W. D. 1984. The attraction of *Zonocerus variegatus* (L) (*Orthoptera*: *Pyrgomorphidae*) to the weed *Chromolaena odorata* and associated feeding behavior. Bulletin of Entomology Research, 74: 239-247.

Ochienga', S. A., Hallberg, E. & Hansson, B. S. 1998. Fine structure and distribution of antennal sensilla of the desert locust *Schistocerca gregaria* (Orthoptera: Acridoidea). Cell and Tissue Research, 291: 525-536.

Muse, W. A. 2003. Effect of sex, age, starvation and feeding, isolation and crowding and oviposition on longevity of Zonocerus variegatus *(L)* (*Orthoptera: Pyrgomorphidae*). Journal of Biological Research and Biotechnology, 1 (2): 11-20.

Tamu, G. 1990. Feeding behavior of the variegated grasshopper *Zonocerus variegates* Ph.D. Thesis, University of Ibadan. 205 pp.

Toye, S. A. 1982. Studies on the Biology of the Grasshopper pest *Zonocerus variegatus (L)* (Orthoptera pyrgomorphia) in Nigeria. Insect Science Application, 3: 1-7.

Wasserman, S. L. & Itagaki, H. 2003. The olfactory responses of the antenna and maxillary palp of the Fleshfly, *Neobellieria bullata* (Diptera; sarcophagidae) and their sensitivity to blockage of nitric oxide synthase. Journal of insect physiology, 49: 271-280.

Youdeowel, A. 1974. Dissection of the variegated grasscopper, *Zonocerus variegatus (L).* Oxford University Press. Ibadan, Nigeria. pp. 69-73.

Zacharuk, R. U. & Shields, V. D. 1991. Sensilla of immature insects. Annual Review of Entomology, 36: 331-354.

Table 1. The measurements of the antenna and the body parts of *Zonocerus variegatus* (n=30).

DEVELOPMENTAL STAGES	ANTENNAL LENGTH (cm)	BODY LENGTH (cm)	BODY WEIGHT (g)	ANTENNAL SEGMENT
1ST INSTAR	$0.19\pm0.1f^{f}$	1.05 ± 0.1^{d}	0.01 ± 0.0^{e}	7.0 ± 0.01^{c}
2ND INSTAR	0.27 ± 0.02^{ef}	1.07 ± 0.25^{d}	0.02 ± 0.01^{de}	9.0 ± 0.01^{bc}
3RD INSTAR	0.29 ± 0.1^{e}	1.15 ± 0.01^{d}	0.03 ± 0.02^{de}	11.0 ± 0.2^{b}
4TH INSTAR	0.45 ± 0.03^{d}	1.53 ± 0.02^{c}	0.05 ± 0.03^{d}	12.0 ± 0.3^{b}
5TH INSTAR	0.54 ± 0.15^{c}	1.96 ± 0.12^{b}	0.14 ± 0.22^{c}	15.0 ± 0.2^{a}
6TH INTAR	0.85 ± 0.01^{b}	2.08 ± 0.1^{b}	0.25 ± 0.14^{b}	16.0 ± 0.5^{a}
ADULT STAGE	1.73 ± 0.04^{a}	3.98 ± 0.01^{a}	0.99 ± 0.05^{a}	17.0 ± 0.2^{a}

Mean values in the same column having the same superscript are not significantly different, $P>0.05$ (SNK)

Table 2: The Ionic Composition of the Antenna of *Zonocerus Variegatus* (mg/mol)

DEVELOPMENTAL STAGES	Ca^{2+}	Mg^{2+}	Zn^{2+}	Na^{+}	K^{+}	Fe^{2+}	PO_4^{2-}	Cl^{-}	TOTAL
1ST INSTAR	0.52 ± 0.1^{b}	0.74 ± 0.01^{a}	0.18 ± 0.3^{a}	2.8 ± 0.1^{b}	14.05 ± 0.1^{c}	0.04 ± 0.0^{d}	4.8 ± 0.2	65 ± 0.5	88.13
2ND INSTAR	1.05 ± 0.1^{a}	0.69 ± 0.01^{a}	0.22 ± 0.1^{a}	3.35 ± 0.6^{a}	16.95 ± 0.0^{a}	0.08 ± 0.1^{cd}	4.3 ± 0.1	61 ± 0.1	87.64
3RD INSTAR	0.26 ± 0.3^{c}	0.14 ± 0.2^{b}	0.11 ± 0.5^{b}	2.66 ± 0.7^{b}	9.1 ± 0.3^{d}	0.09 ± 0.0^{cd}	3.2 ± 0.1	40 ± 0.0	55.56
4TH INSTAR	0.26 ± 0.2^{c}	0.24 ± 0.5^{b}	0.11 ± 0.6^{b}	1.4 ± 0.2^{d}	0.38 ± 0.1^{f}	0.12 ± 0.2^{bc}	4.0 ± 0.3	60 ± 0.0	66.51
5TH INSTAR	0.22 ± 0.7^{c}	0.27 ± 0.1^{b}	0.09 ± 0.2^{b}	1.9 ± 0.1^{c}	5.4 ± 0.1^{e}	1.14 ± 0.1^{a}	3.5 ± 0.4	45 ± 0.1	57.52
6TH INSTAR	0.22 ± 0.2^{c}	0.6 ± 0.5^{a}	0.1 ± 0.6^{b}	1.2 ± 0.6^{d}	14.94 ± 0.2^{b}	0.17 ± 0.5^{b}	2.6 ± 0.1	32 ± 0.8	51.83
ADULT STAGE	0.19 ± 0.1^{c}	$0.23\pm^{b}$	0.09 ± 0.8^{b}	1.9 ± 0.1^{c}	4.96 ± 0.1^{e}	0.19 ± 0.6^{b}	3.8 ± 0.4	58 ± 0.6	69.36

Mean values in the same column having the same superscript are not significantly different, $P>0.05$ (SNK)

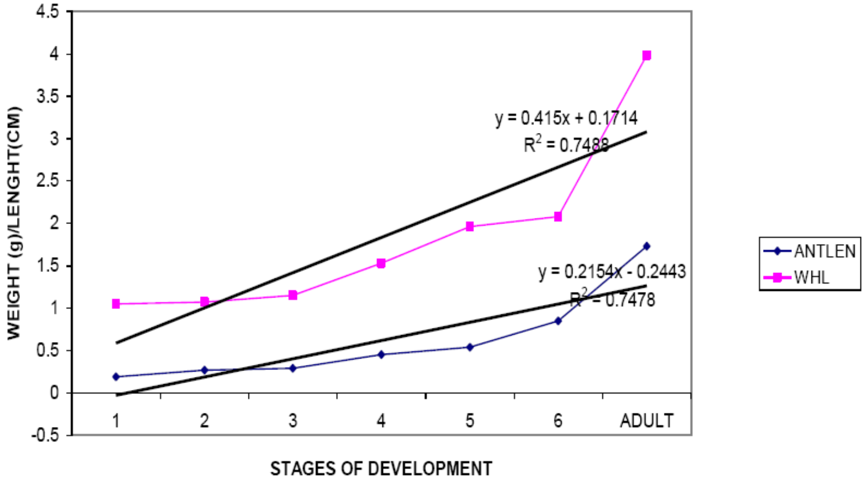

Figure 1. Growth pattern of the antennae and body length of *Z. variegatus* (L.).

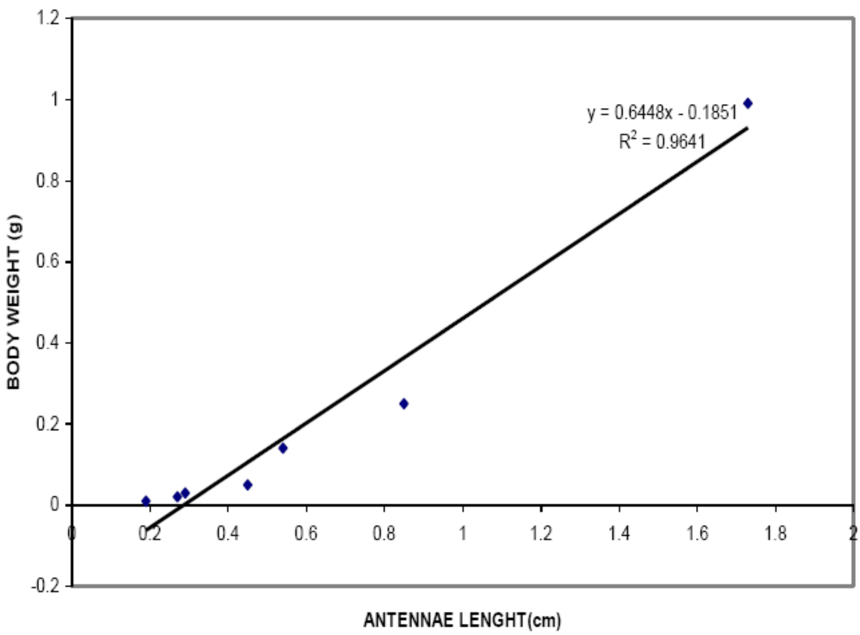

Figure 2. Regression (r2) between the antennal length and body weight of *Z. variegatus* (L.)

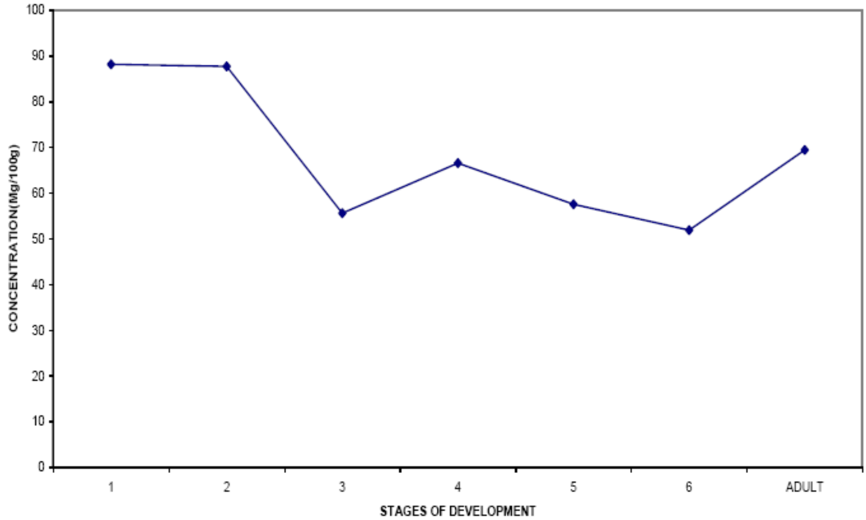

Figure 3. Total ionic composition of antennae of *Z. variegatus* (L.)