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Do nanoparticles cause hormesis? Early physiological compensatory response in house crickets to a dietary admixture of GO, Ag, and GOAg composite

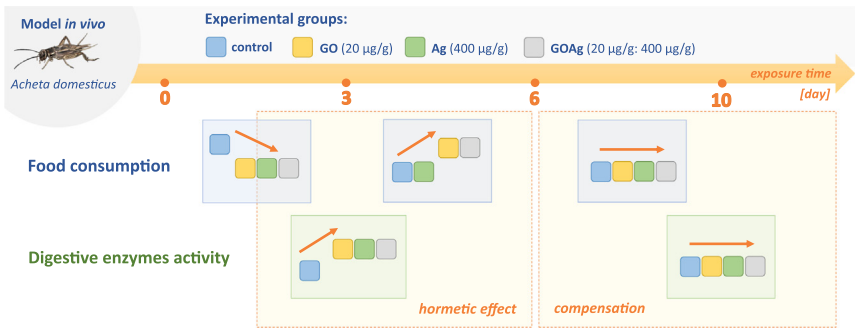
Reyhaneh Seyed Alian [a], Marta Dziewięcka [a], Andrzej Kędziorski [a], Łukasz Majchrzycki [b], Maria Augustyniak [a,*]

[a] Institute of Biology, Biotechnology and Environmental Protection, Faculty of Natural Sciences, University of Silesia in Katowice, Bankowa 9, 40-007 Katowice, Poland
[b] Centre of Advanced Technology, Adam Mickiewicz University in Poznan, ul. Uniwersytetu Poznańskiego 10, 61-614 Poznań, Poland

HIGHLIGHTS

- Low amounts of GO, AgNPs, or GO-AgNPs triggers an early stress response in crickets.
- Nanoparticles (GO, AgNPs, or GO-AgNPs) can disturb the energy budget of crickets.
- Initial suppression of energy consumption/assimilation is compensated afterwards.
- Increased activity of digestive enzymes accompanies the compensation response.
- Insects treated with GO-AgNPs composite retained more body water.

GRAPHICAL ABSTRACT



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ABSTRACT

This study aimed to identify the physiological responses of house cricket females following short-term exposure to relatively low dietary doses of graphene oxide (GO, 20 μg · g^{-1} food), silver (Ag, 400 μg · g^{-1} food) nanoparticles (NPs), or graphene oxide-silver nanoparticle composite (GO-AgNPs, 20: 400 μg · g^{-1} food). Energy intake and distribution were measured on the third, sixth, and tenth day. A semi-quantitative API®ZYM assay of digestive enzyme fingerprints was performed on the third and tenth day of continuous treatment. Physicochemical properties of the NPs were obtained by combining SEM, EDX spectrometry, AFM, and DLS techniques.
The obtained results showed decreased energy consumption, particularly assimilation as an early response to dietary NPs followed by compensatory changes in feeding activity leading to the same consumption and assimilation throughout the experimental period (10 days). The increased activities of digestive enzymes in NP-treated females compared to the control on the third day of the experiment suggest the onset of compensatory reactions of the day. Moreover, the insects treated with GO-AgNP composite retained more body water, suggesting increased uptake. The observed changes in the measured physiological parameters after exposure to NPs are discussed in light of hormesis.

1. Introduction

Recent years have brought the dynamic development of nanotechnology and an exponential increase in the synthesis of new nanoparticles. Silver nanoparticles (AgNPs) and graphene (its derivatives

* Corresponding author.
E-mail address: maria.augustyniak@us.edu.pl (M. Augustyniak).

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including graphene oxide (GO)) are widely used nanoparticles with significant potential. Their specific physicochemical properties allow them to be suitable for a wide array of applications (Wu et al., 2012; Dideikin and Vul, 2019; Smith et al., 2019; de Medeiros et al., 2021). Silver nanoparticles, due to their unique optical, catalytic, photo-thermal, and antibacterial properties, are currently the most commonly used nanoparticles (Rai et al., 2009; Taglietti et al., 2012; Katz et al., 2015; Wei et al., 2015; Pulit-Prociak and Banach, 2016; McGillicuddy et al., 2017).

The synthesis of hybrid materials, for example, GO-metal nanocomposites, aims to enhance their functionality, provide novel properties, and consequently, new applications (Zhu et al., 2013; Bhunia and Jana, 2014; de Saravia et al., 2020; de Medeiros et al., 2021). GO and AgNPs are considered to work synergistically, and the GO-AgNP nanocomposite may have better antimicrobial and catalytic activities and higher thermal conductivity than its components (Tang et al., 2013; Cobos et al., 2020). Hence, GO-AgNP composites are of great interest and potential, including anticancer therapy (Gurunathan et al., 2015; Kavinkumar et al., 2017). However, the antibacterial properties of GO, AgNPs, and GO-AgNP composites should be considered due to their effects on microorganisms that constitute the intestinal or gut microbiome (Li et al., 2018). Accidental contamination of the environment/food with such material can lead to impairment of digestive functions, indirectly affecting the nutrients and energy intake and, consequently, the energy budget of an organism.

The production and application of nanoparticles, including GO, Ag, and AGO-Ag composites, is constantly increasing (Pulit-Prociak and Banach, 2016; Inshakova and Inshakov, 2017; Malakar A et al., 2021). Therefore, they can be common environmental contaminants to which innumerable organisms could be exposed. The problem of the unfavorable influence of nanoparticles often manifested in the inhibition/impairment of various physiological processes, is frequently investigated. Less attention has been given to the potential hormetic/compensatory effects of nanoparticle exposure.

Hormesis consists of a two-phase reaction of an organism to a stressor (a chemical compound or an environmental factor) - low doses cause stimulation or beneficial effects, and high doses inhibit numerous processes and cause unfavorable effects (Mattson, 2008). Hormesis is an adaptive compensatory response that follows the initial disturbance of an organism's homeostasis. It is worth considering that hormetic effects are usually subtle/modest changes, which sometimes makes it difficult to distinguish from natural variability within a specific parameter (Hoffmann and Stempsey, 2008; Wang et al., 2017). One of the first hormesis studies was performed on house cricket, *Acheta domesticus* (Luckey, 1968; Cohen, 2005; Guedes et al., 2009; Cutler, 2013).

Toxicity studies on nanoparticles continuously provide new data. However, few studies have focused on doses/concentrations below the toxic threshold. In some studies that have focused on the biological effects of low doses of nanoparticles a hormetic effect has been observed. There are examples of in vitro studies that used carbon nanotubes, nanodiamonds, quantum dots, or metal nanoparticles, mostly silver. A few in vivo studies have described hormetic for various/specific endpoints as an effect of exposure to metal nanoparticles or carbon nanotubes. These studies were performed on selected strains of bacteria and some aquatic biota, including some species of algae and plants, few crustaceans, and few vertebrates (Iavicoli et al., 2010; Iavicoli et al., 2018; Agathokleous et al., 2019). However, the data on this phenomenon, particularly regarding insects and the potential energy costs of the supposed compensation, are limited and very fragmented or even missing.

Changes caused by additional stress factors should be accompanied by an increase in the energy consumption to activate defense, repair, and/or compensatory mechanisms. Increased energy demand should require an increase in food consumption. If the conditions are favorable, an increase in the efficiency of the digestive system is needed. Hence, estimating the energy budget and examining the overall efficiency of digestive enzymes is strongly justified.

The basic parameter of the animal's energy budget is consumption (C). Assimilation (A) is a part of the energy absorbed by the organism that is spent on respiration (R) to maintain life processes and production (P) related to weight gain, body reconstruction, and reproduction. In insects, energy allocation to production is associated with critical stages of development, molting, cocoon formation, and egg production (Schowalter, 2006; Gao et al., 2007; Yates et al., 2011). Adequate energy flow through an organism and its distribution is a basic demand for life, and various stressors may disturb its intake and allocation. However, sufficiently low “stressors' doses” might cause a compensatory response that could increase the chances of coping with unfavorable conditions.

In previous long-term studies on NP effects in crickets, we found mostly toxic effects of varying severity on different tissue/cellular/molecular endpoints (Dziewięcka et al., 2015; Karpeta-Kaczmarek et al., 2016a–c; Dziewięcka et al., 2017; Dziewięcka et al., 2018; Karpeta-Kaczmarek et al., 2018; Dziewięcka et al., 2020: Flasz et al., 2020). During our observations, particularly in the initial stage of exposure, we observed an increased consumption, compared to the control, in crickets provided with food contaminated with low concentrations of NPs (e.g., 20 μg GO/g of food), with no apparent toxic effects at the systemic level. Simultaneously, at high concentrations (e.g., 200 μg GO/g of food), food consumption contaminated with nanoparticles decreased significantly compared with the control group (unpublished data). These observations encouraged us to design an experiment to assess the detailed energy budget and digestive enzymes in the first 10 days of exposure to nanoparticles: GO, Ag, and GO-Ag composite. As we were interested in changes over time (i.e., multiple measurements within the experimental group), we chose only one concentration for each NP that seemed to cause increased consumption in our previous studies. Thus, this study aimed to quantify the expected changes and check whether the changes in food intake and utilization are accompanied by changes in the efficiency of the digestive system. Therefore, we evaluated the intestinal enzyme fingerprints (Kaufman et al., 1989; Boetius and Felbeck, 1995; Doherty-Weason et al., 2019) and the energy budget of *A. domesticus*. We hypothesized that nanoparticles could change gut function and deregulate the energy budget. However, considering the low doses used in this study and the short exposure time, possible activation of compensatory mechanisms could restore the measured parameters to the level of unexposed individuals.

2. Materials and methods

2.1. House cricket

Acheta domesticus (Orthoptera, Insecta) is often used as a model organism in physiological and toxicological research (Szelei et al., 2011; Horch et al., 2017). The species is omnivorous and easy to breed; the life cycle takes approximately 2–3 months. The animals used in the experiment were obtained from our laboratory stock colony.

2.2. Nanoparticles selection

Ag, GO nanoparticles, and their GO-Ag composite were used as exposure factors; their effect on the body, due to their properties, may have a different mechanism. The GO-Ag composite was prepared as a separate nanoparticle solution with a concentration ratio of the working components (GO:Ag = 20:400). This approach allowed the observation of the potential enhancement of the AgNP performance after anchoring to GO sheets or the additive effect of the two types of components. The specific structure of the nanocomposite can provide a unique nanointerface for interaction with the gut microbiome and facilitate the interaction between AgNPs and GO sheets, resulting in a synergistic effect (Tang et al., 2013).

2.3. Preparation of graphene oxide suspension, silver suspension, and Ag-GO composite

Graphene oxide powder (15–20 sheets, Sigma Aldrich) was sonicated in deionised water (10 mL) by ultrasonic homogenizer (cycle: 1, amplitude 100%; model UP-100H, DONSERV) until a homogeneous GO suspension (10 mg/mL).

Silver nanoparticles (99.9%, 20–30 nm, SS Nanomaterials, Inc.) were dispersed in a citrate buffer solution (0.1 M, 10 mL, pH =6.5) by sonication (cycle: 1, amplitude 100%) to form a stable Ag colloid solution (10 mg/mL).

Graphene oxide-silver nanoparticle composite suspension (GO-AgNPs) was obtained due to the mixing of silver nanoparticles with graphene oxide in a 5:1 ratio deionised water and citrate buffer solution (0.1 M; pH = 6.5). Briefly, 2 mg of graphene oxide powder was dispersed in 100 mL (20 ppm) deionised water followed by sonication for 2 h to greater homogeneity. Thereafter, silver colloid in the 0.1 M citrate buffer (400 ppm, 20 mL) was added to the GO suspension and gently heated while sonication for 1 h. The final product was left overnight in the dark at room temperature (Bao et al., 2011; Dinh et al., 2014; Xue-Fei et al., 2015).

2.4. NP characterization

The morphology and structure of the Ag, GO nanoparticles, and GO-AgNP composites were characterized using a scanning electron microscope (SEM) with an energy dispersion X-ray spectrometer (EDX) (Quanta FEG 250; FEI) and atomic force microscopy (AFM)(Agilent 5500). The size distribution profile of the Ag nanoparticles was determined by dynamic light scattering (DLS) equipped with 4 mW He-Ne, 633 nm Malvern Zetasizer Nano-ZS (Malvern Instruments, U.K.).

Samples for analysis were highly diluted and deposited on a silicon wafer (SEM) and fresh cleaved mica for AFM measurements. SEM imaging was performed with a beam accelerating voltage of 2 kV under high-vacuum conditions to ensure good contrast during imaging. Energy-dispersive X-ray spectrometry was used to identify and quantify the elemental composition of the GO-AgNP composite. EDX spectra from individual particles were analyzed using a vector-based algorithm to determine the elements. The AFM measurements were performed in tapping mode with a typical force constant of 40 N/m and a resonant frequency of 350 kHz. The standard scan frequency was 0.2. The DLS measurements were taken at 21 °C for 200 s with a 173° detection angle. The final results were calculated using the size distribution by the number technique.

The results showed well-defined GO structures, mostly single-layered with a typical height of 1.0 nm and an average flake area of approximately 2 μm (Fig. 1 A2, B2). Silver nanoparticles were present as aggregates with a diameter of about 65 nm (Fig. 1 B1) and individual particles in the range of 5–20 nm (Fig. 1 B2). The GO-AgNP composite was in the form of thick GO aggregates between and 5–40 nm coated with silver nanoparticles (Fig. 1 A3, B3, D1).

2.5. Experimental set-up for energy budget and digestive enzyme screening

Adult females (0–1 d after final molting) were randomly divided into control, and three experimental groups provided feed supplemented with NPs: GO (graphene oxide at 20 μg/g of food), Ag (silver nanoparticles, 400 μg/g of food), and GOAg (20 and 400 μg of GO and AgNPs, respectively, per gram of food). Each group consisted of six sub-groups kept in plastic boxes (28 × 20 × 16 cm; 7 individuals in each) under standard conditions (28.8 °C ± 0.88 °C, 20%–45% RH, and photoperiod L:D 12:12). A weighed amount of feed was provided on days 0, 3rd and 6th day (Dziewięcka et al., 2017). The remains and feces were collected separately on 3rd, 6th and 10th day. The increase in female body weight was measured with 1 mg accuracy on days 0, 3rd, 6th and 10th (final). All the collected feed and feces samples were dried at 40 °C for 24 h to obtain their dry weight. The 10th-day females were anesthetized with CO_2, then frozen and lyophilized for approximately 44 h. An additional reference group of 0-day females was frozen and lyophilized to further calculate the initial dry weight of females from the groups.

Separate plastic boxes (2 for each experimental group: control, GO, Ag, and GOAg) were set up to analyze digestive enzyme activity. The insects were fed ad libitum with the same food and kept under the same conditions as for calculation of the energy budget. Each plastic box contained ten 0–1-day-old females. Insects for the digestive enzyme activity assay were collected on the third and tenth day of adult age.

2.6. Calorimetry assay

Samples of the feed and feces were re-dried in a halogen moisture analyzer (HR73, Mettler Toledo, Switzerland) immediately before preparation of a 100 mg pellet for combustion in a 6725 semi-microcalorimeter (Parr Instrument Company, USA) under high oxygen pressure (2.8 MPa). After the combustion of the sample, the vessel was washed with distilled water, and the washing was titrated with 0.0709 N Na_2CO_3 using methyl red. Before sample determination, the device was calibrated using benzoic acid as a thermochemical standard.

Females were combusted in the same way. The unknown calorific value (CV) of the female samples was calculated from their dry weight and an appropriate regression equation ($R^2 = 0.991$).

2.7. Energy budget calculations

Consumption and assimilation rates and approximate digestibility (AD) were calculated for particular intervals and the entire experimental time. For the latter, the growth rate and efficiency of ingested energy conversion into bodyweight (ECI) and efficiency of digested energy conversion into bodyweight (ECD) indices were calculated according to standard formulas (Waldbauer, 1968).

2.8. Enzymatic activities screening by API® ZYM system

Enzymatic activity in the gut was assayed using an API®ZYM kit (BioMérieux, France) designed for cell suspensions and successfully used in assaying enzymes for invertebrate species (Kaufman et al., 1989; Boetius and Felbeck, 1995; Collin and Starr, 2013; Doherty-Weason et al., 2019; Przemieniecki et al., 2020). The API®ZYM test can be used as a general (screening) method of assessing an organism's digestive capacity, concerning the total activity of digestive enzymes derived from microbiota and their host, that is, *A. domesticus* in this case.

The API®ZYM strip test contains dehydrated chromogenic substrates of 19 enzymatic reactions involved in the breakdown of lipids, peptides, phosphoric esters, and polysaccharides. The enzymes and substrates included in this study are listed in Table 1.

A cell suspension of the required protein concentration was prepared for each experimental group by homogenizing the midgut dissected from three ice-anesthetized females. The same mass of intestinal fragments (75 ± 2 mg) was collected to obtain a homogenate with the required volume needed for the test and a similar protein concentration. The homogenates were centrifuged at 10,000 rpm for 10 min at 4 °C.

According to the manufacturer's protocol, the API test plates were placed in moist chambers, and 65 μL of supernatant was added to each well, followed by incubation for 3 h at 37 °C in the dark. After incubation, reagents Zym A and Zym B were added to stop the reaction, and the resulting color was allowed to develop for 5 min. Any excess of unreacted Fast Blue BB was removed by short-term exposure to a strong light source (1000 W, 10 s, 10 cm from the plate), and after aligning the colors, the plates were photographed.

The semi-quantitative assay was based on a visual comparison of the colors on the test strips obtained by enzymatic reactions with a

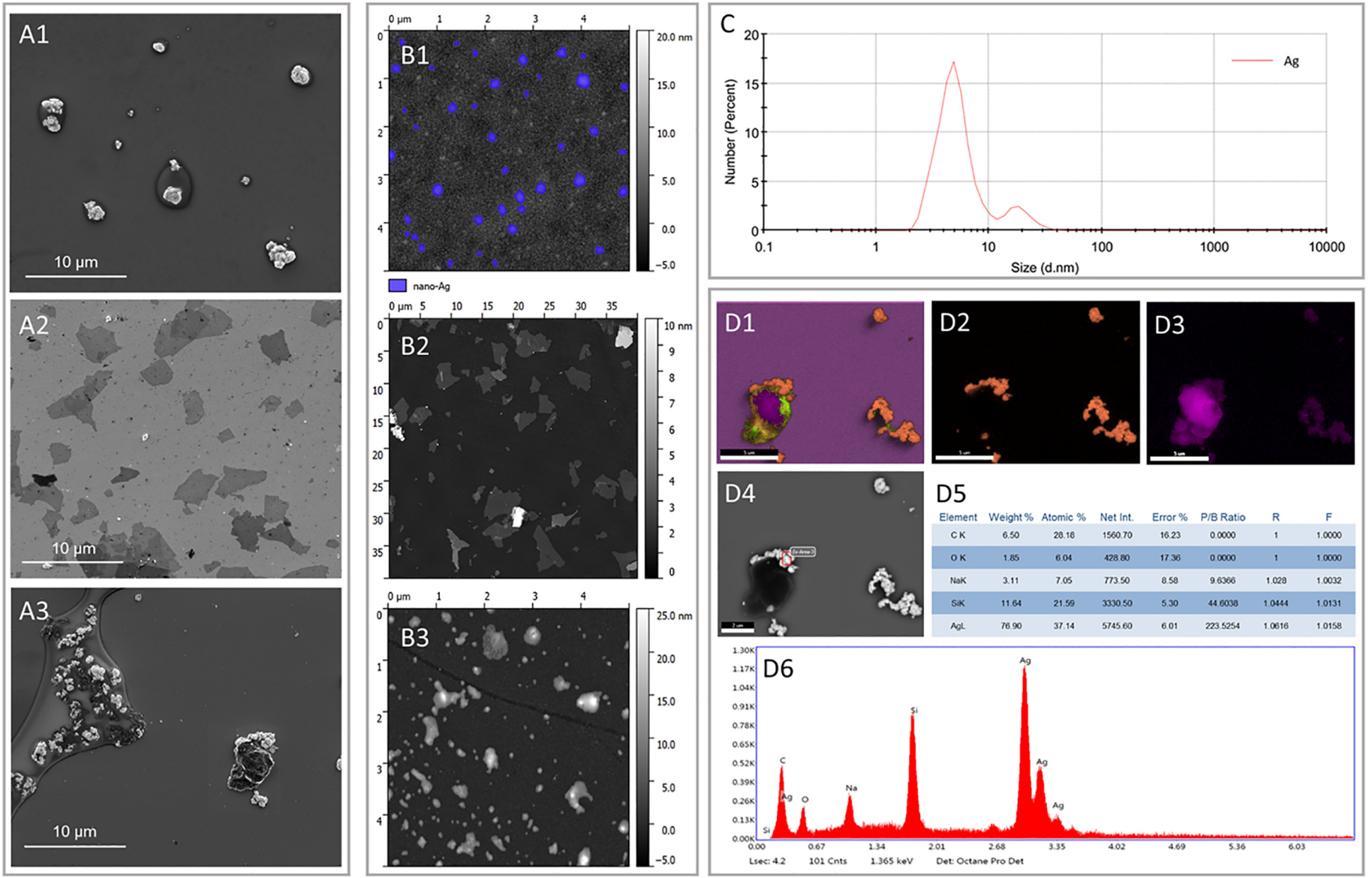

Fig. 1. Physicochemical characteristics of nanoparticles. (A) SEM images of silver nanoparticles (A1), graphene oxide (A2), GO-AgNPs composite (A3). (B) AFM images of silver nanoparticles (B1), graphene oxide (B2), GO-AgNPs composite (B3). (C) Dynamic light scattering (DLS) of silver nanoparticles. (D) SEM-EDS elemental mapping of GO-AgNPs composite (D1) with representative particles: Ag (D2), and carbon (D3). Spot EDS of GO-AgNPs (D4) with table for the atomic and weight percentage of various elements (D5) and area EDS spectrum (D6).

Table 1
Enzymes and substrates in API ZYM test (BioMérieux).

No.	Enzyme	APIZYM substrates	Abbreviation
1	*Negative control*	None	
2	*Alkaline phosphomonoesterase*	2-Naphtyl phosphate	AlP
3	*Esterase (C4)*	2-Naphtyl butyrate	Est
4	*Esterase lipase (C8)*	2-Naphtyl caprylate	EstLip
5	*Lipase*	2-Naphtyl myristate	Lip
6	*Leucine-arylamidase*	L-Leucyl-2-naphthylamide	ALeu
7	*Valine arylamidase*	L-Valyl-2-naphthylamide	AVal
8	*Cystine arylamidase*	L-Cystyl-2-naphthylamide	ACys
9	*Trypsin*	*N*-Benzoyl-DL-arginine-2-naphtylamide	T
10	*α-chymotrypsin*	*N*-Glutatyl-phenylalanine-2-naphthylamide	ChT
11	*Acid phosphatase*	2-Naphthyl phosphate	AcP
12	*Naphthol-AS-BI-phosphohydrolase*	Naphthol-AS-BI-phosphate	NPH
13	*α-Galactosidase*	6-Br-2-naphthyl-αD-galactopyranoside	αGal
14	*ß-Galactosidase*	2-Naphthyl-βD-galactopyranoside	ßGal
15	*ß-Glucuronidase*	Naphthol-AS-BI-βD-glucuronide	ßGlr
16	*α-Glucosidase*	2-Naphthyl-αD-glucopyranoside	αGlu
17	*ß-Glucosidase*	6-Br-2-naphthyl- βD-glucopyranoside	ßGlu
18	*N-acetyl-ß-glucosaminidase*	1-Naphthyl-*N*-acetyl-βD-glucosamide	NAG
19	*α-Mannosidase*	6-Br-2-naphthyl- αD-mannopyranoside	αMan
20	*α-Fucosidase*	2-Naphthyl-αL-fucopyranoside	αFuc

manufacturer's standard. The enzymatic activity (color intensity of the wells) was measured using ImageJ® software using grayscale images. Each enzyme was described as the arithmetic mean of the frequency plot (1D histogram) over the range [0.255] and converted relative to the control well.

2.9. Statistical analysis

The measurements were performed in six replicates. The distribution of the data and homogeneity of variance were checked for all parameters before analysis. The Kolmogorov-Smirnov and Lilliefors tests for normality and Levene's test for variance homogeneity allowed the analysis of variance. As the data fulfilled the analysis of variance assumptions, we performed parametric tests to check for differences between experimental groups. The least significant difference test (ANOVA, LSD test, $p < 0.05$) was performed separately for each parameter and each time point. All parameters are expressed as mean + SE in the figures. Statistical analyses were performed using the Statistica 13.1.

3. Results

3.1. Energy budget

Ten-day-lasting exposure of young female crickets to nanoparticles (NPs) revealed time-dependent changes in food (energy) consumption and utilization.

The average daily consumption per individual was approximately 800 J in all groups except females from the GO group that ingested 24% more energy than the control insects (Fig. 2B). The highest daily consumption in all groups occurred during the first 3 days of the experiment; however, it was significantly lower in the GOAg group's crickets.

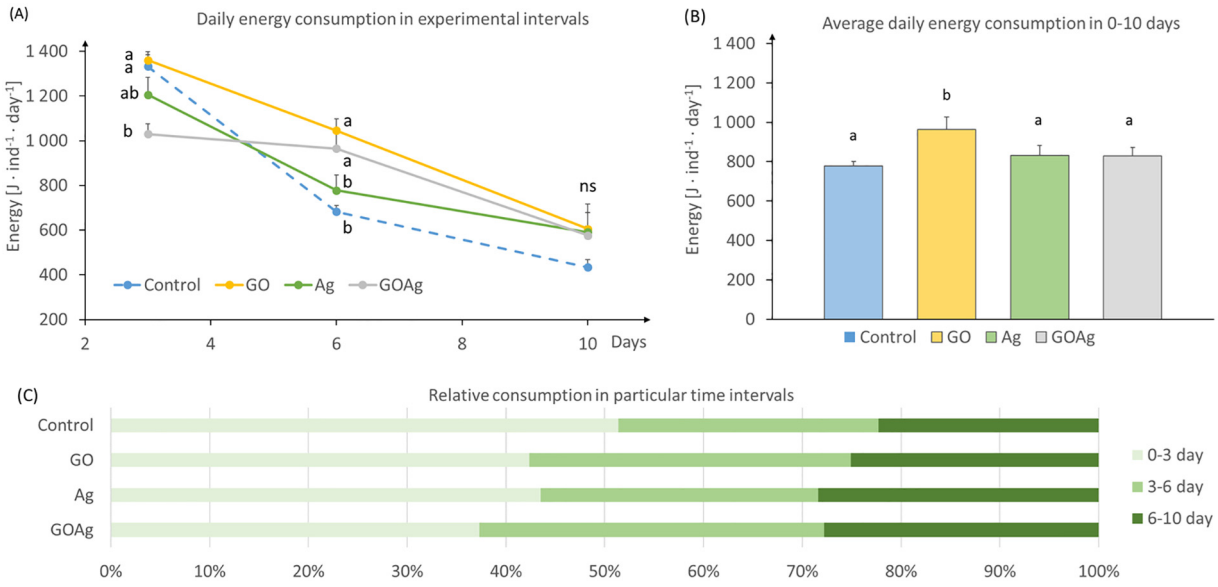

Fig. 2. Daily energy consumption (mean + SE) by young female crickets during the subsequent time intervals (A), the whole experiment (B), and its changes among the intervals (C). The same letter denotes homogenous group (ANOVA, NIR, $p < 0{,}05$). Experimental groups: GO – crickets exposed to graphene oxide admixture at 20 μg · 1 g^{-1} dry weight feed, Ag – crickets exposed silver nanoparticles at 400 μg · 1 g^{-1} dry weight feed, GOAg – crickets exposed to both GO and AgNPs in the given amounts; experimental intervals: 0–3 days, 3–6 days, and 6–10 days.

In the following days, the energy intake decreased, although it was higher in the NP-treated groups than in the control group (Figs. 2A,C). However, during the 6–10 d period, the differences between the experimental groups were not significant.

Average energy assimilation during the whole experiment did not differ among the experimental groups; however, the differences occurred in the subsequent measurement time intervals (Figs. 3A-C). Energy assimilation followed the consumption "pattern," however it was significantly lower in all NPs-treated groups than in control during the first 3 days. The control females assimilated 52.3% of the total assimilated energy in this interval, whereas the NPs-exposed, up to 40% and 37.8% in the Ag and the GOAg groups, respectively (Figs. 3A, C). In the next 3 days, the decrease in assimilation was much lower in the GO and GOAg groups than in the control and Ag groups. After the sixth day, the assimilation in the groups receiving nanoparticles did not differ from that in the control group (Fig. 3A).

The approximate digestibility of ingested energy was significantly lower in NP-exposed crickets (up to 20% in the GO group) than in the control ones during the first 3 days of the experiment. This difference diminished in the following days; therefore, digestibility in the whole experimental period in the NP-treated crickets was only 4%–7% lower than that in the control (Figs. 4A-B). Calculations of ECD values revealed similar efficiency of digested energy conversion into the tissues (28.2%–22.2%). This means that approximately 70% of the assimilated energy was allocated to maintenance costs.

Comparison of water content in the samples revealed that feces of NP-treated crickets contained more H_2O than feces of the control ones by 25%–30% in the 0-3rd day period, and 14.7%–26.5% in the 3th–6th day period. Only the feces of the GOAg crickets had higher water content than that of the control (Fig. 4C). Interestingly, females from this group also retained more water in their tissues than did the control insects (Fig. 4D).

3.2. Gut enzymes' activities

Following quantitative transformation of the API®ZYM test, we observed time-dependent changes in the activity of gut enzymes. The highest relative activity was measured for enzymes digesting carbohydrates, whereas proteolytic and esterolytic enzymes were less important. This difference may reflect the food composition mainly containing carbohydrates, including fiber.

On the third day of the experiment, we observed the increased activity of most assayed carbohydrate-degrading enzymes in NP-exposed crickets, particularly those hydrolyzing β-glycosidic bonds such as β-galactosidase and β-glucosidase. Generally, the highest stimulation of these enzymes versus control was observed in females from the Ag group, while the lowest was observed in females from the GOAg group. The activity of α-mannosidase was lower following NP exposure, and α-galactosidase showed the lowest activity in all the groups (Fig. 5A).

On the 10th day, this stimulatory effect disappeared in all the assayed enzymes, and in the case of β-glucosidase and *N*-acetyl-β-glucosaminidase activity we measured slight inhibition (Fig. 5B).

Both acid (AcP) and alkaline (AlP) phosphatase and naphthol-AS-BI-phosphohydrolase (NPH) activity were higher in NP-treated female crickets than in the control on the third day of exposure (Fig. 5C). At the end of the experiment, the activity was the same in all the groups (Fig. 5D).

We also observed an elevated proteolytic activity on the third day of NP treatment compared to the control, particularly in the case of arylamidases. Higher activity of trypsin was observed only in the GO group (Fig. 6A). On the 10th day, this effect disappeared, and no stimulation or inhibition was observed (Fig. 6B).

The activity of esterolytic enzymes followed the same 'pattern' – stimulated activity in exposed crickets was observed on the third day and the same activity in all the groups – on the 10th day of the experiment.

4. Discussion

Hormetic factors (stressors, also changes in energy availability) trigger a stress reaction in the cell/organism, which is manifested, for example, by changes in the concentration of free radicals, disturbed ion distribution, increased consumption of energy reserves, and others. The most frequently described hormetic effects in response to this moderate stress are increased defense strategies, for example, enhanced activity of antioxidative enzymes, stimulation of chaperones indicating the intensification of protein synthesis/destruction processes,

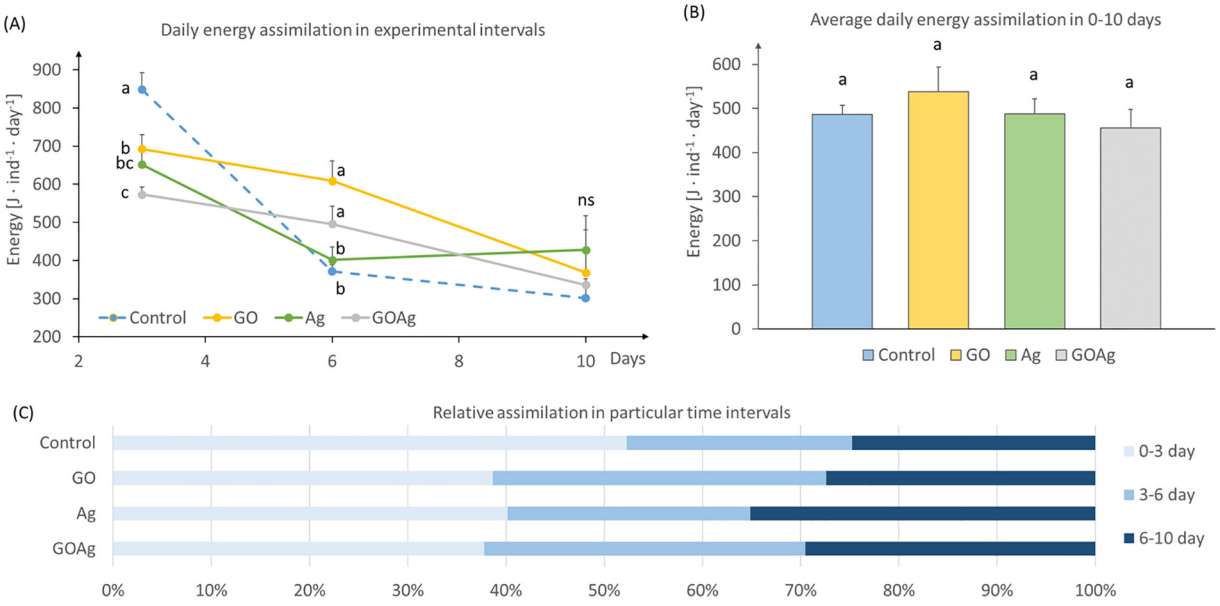

Fig. 3. Daily energy assimilation (mean + SE) by young cricket females during the subsequent time intervals (A), the whole experiment (B), and its changes among the intervals (C). The same letter denotes homogenous group (ANOVA, NIR, $p < 0,05$). Description of the groups as in Fig. 2.

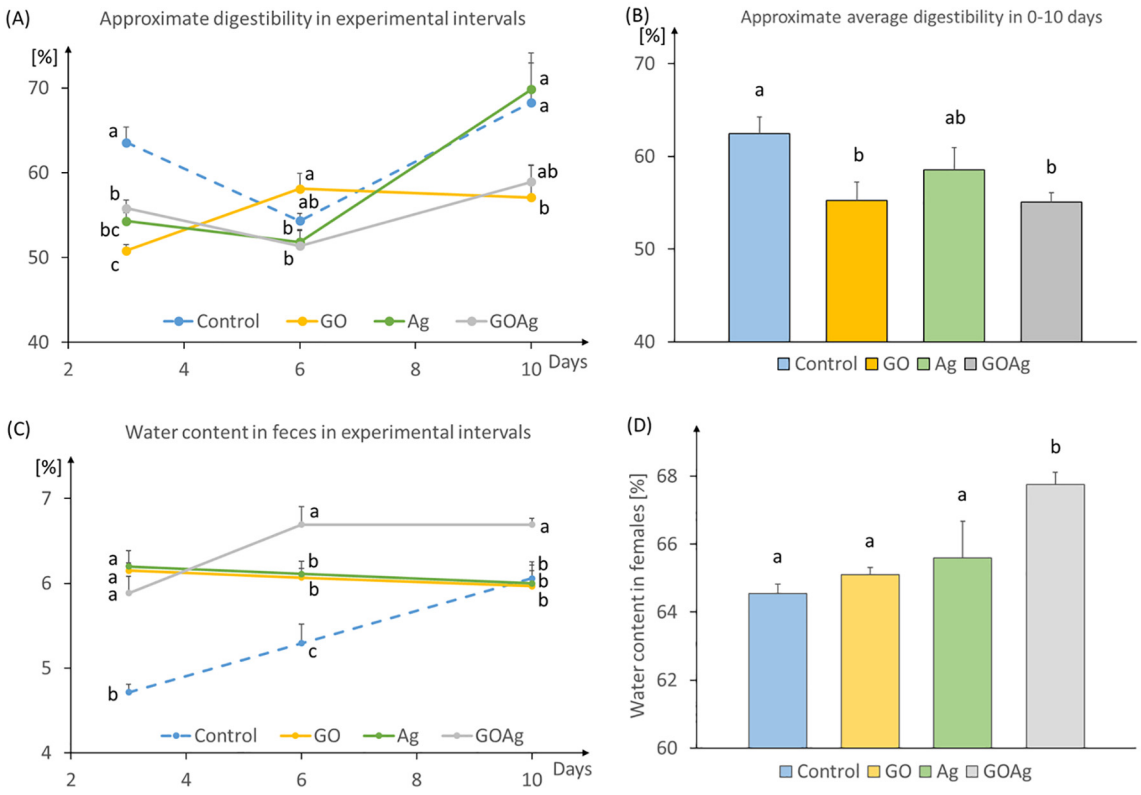

Fig. 4. Digestibility of ingested energy in subsequent time intervals (A), the whole experiment (B), and water content (mean + SE) in the feces (C) and females at the end of the experiment (D). The same letter denotes homogenous group (ANOVA, NIR, $p < 0,05$). Description of the groups and experimental intervals as in Fig. 2.

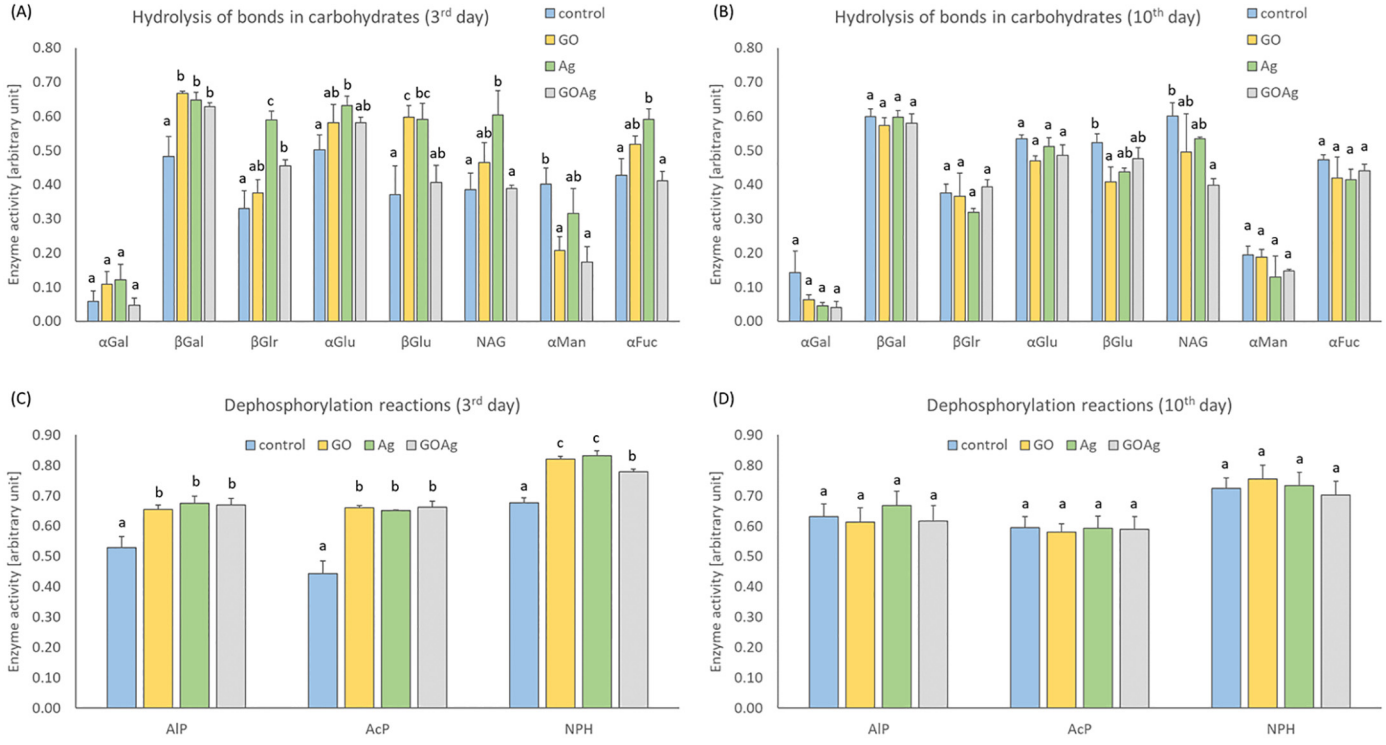

Fig. 5. Relative activity (mean + SE) of glycosidic bond-clevaging (A and B) and dephosphorylating (C and D) enzymes in digestive tract of house cricket females from control and nanoparticles-treated groups at 3rd (A, C) and 10th (B, D) day of continuous treatment. The same letter denotes homogenous group for particular enzyme (ANOVA, NIR, $p < 0,05$). Abbreviations of the groups as in Fig. 2; Glycolytic enzymes: αGal - α-galactosidase, βGal - β-galactosidase, βGlr - β-glucuronidase, αGlu – α-glucosidase, βGlu – β-glucosidase, NAG – *N*-acetyl-β-glucosaminidase, αMan – α-mannosidase, αFuc – α-fucosidase; Dephosphorylating enzymes: AlP – Alkaline phosphatase, AcP – Acid phosphatase, NPH – Naphthol-AS-BI-phosphohydrolase.

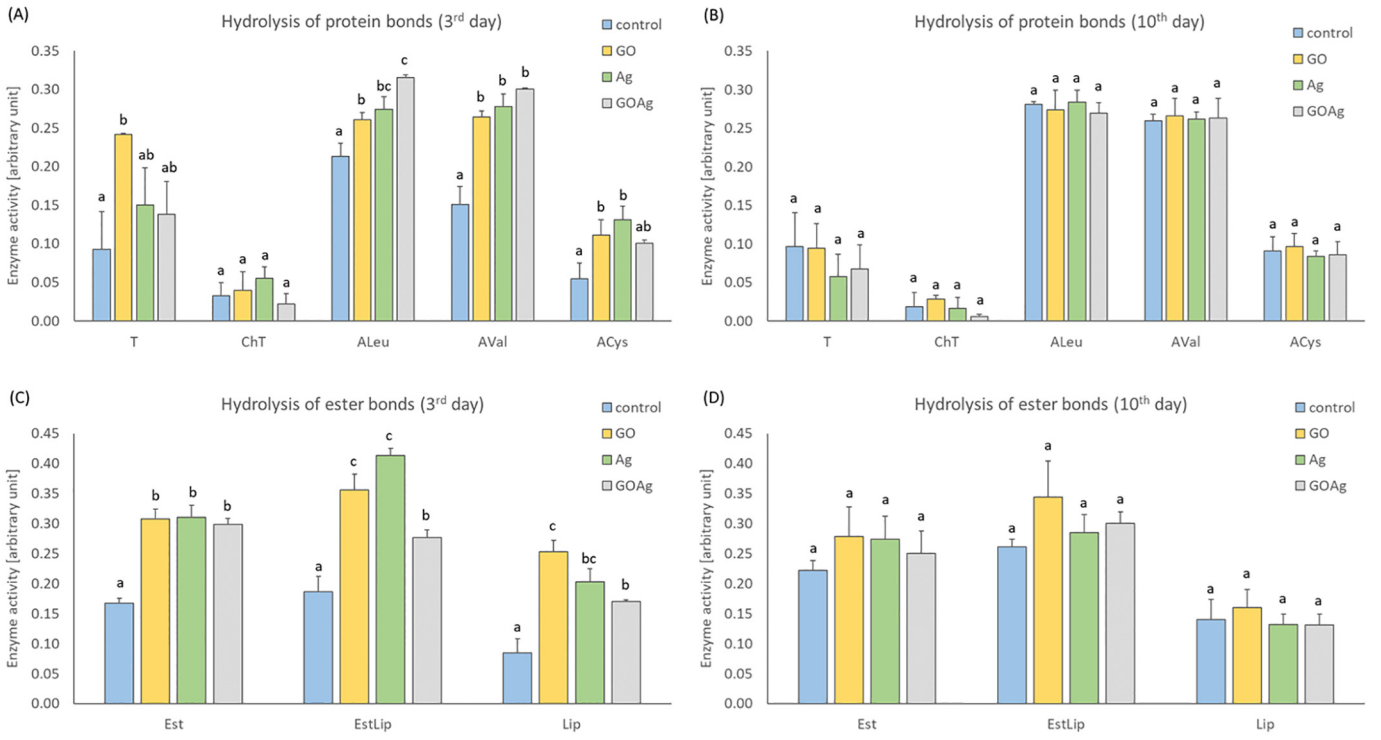

Fig. 6. Relative activity (mean + SE) of proteolytic (A and B) and esterolytic (C and D) enzymes in digestive tract of house cricket females from control and nanoparticles-treated groups at 3rd (A, C) and 10th (B, D) day of continuous treatment. The same letter denotes homogenous group for particular enzyme (ANOVA, NIR, $p < 0{,}05$). Experimental groups: GO – crickets exposed to graphene oxide admixture at 20 μg · 1 g^{-1} dry weigh feed, Ag – crickets exposed silver nanoparticles at 400 μg · 1 g^{-1} dry weigh feed, GOAg – crickets exposed to both GO and Ag in the given amounts; Proteolytic enzymes: T – trypsin, ChT – α-chymotrypsin, ALeu – Leucine arylamidase, Aval – Valine arylamidase, ACys – Cystine arylamidase; Esterolytic enzymes: Est – Esterase (C4), EstLip – Esterase Lipase (C8), Lip – Lipase (C14).

stimulation of growth factors, and proteins involved in energy metabolism. All these factors protect the cell/organism from more severe stress (Mattson, 2008). However, it should be noted that these defense processes require additional energy, which may increase food consumption and assimilation and presumably improve the digestive system's efficiency.

This study found that nanoparticles present in the food could temporarily disturb the cricket's energy budget and change digestive enzyme activities (Figs. 2–6). During the first exposure period (0–3 days), a significant reduction in consumption and assimilation was observed (Figs. 2A and 3A), which can be a manifestation of the initial disturbance of homeostasis (Calabrese, 2005). However, the analysis of the fingerprints of the gut enzymes on the third day of exposure (Figs. 5A and C, Figs. 6A and C) showed that the compensation mechanisms were triggered. They may be seen as a smaller drop in consumption and assimilation during the following intervals in NP-treated crickets than in control ones (i.e., days 3–6; Figs. 2 and 3 - see GO and GOAg groups). Higher energy intake and absorption than in the control might be a hormetic response triggered by low "pressure" of stressors. This suggests that the nanoparticle concentrations in our experiment were low enough to cause such a hormetic effect. Taking into account the concentration of NPs in the food and the consumption rate, we calculated that in the first 3 days of exposure, *A. domesticus* could take up 4.607 μg GO (GO group), 81.617 μg Ag (Ag group), and 3.488 μg GO and 69.762 μg Ag (GOAg group) with food. In fact, the biological effects of NPs have been studied over a wide range of concentrations, and the amounts used in the present work are among the lowest (Guo and Mei, 2014; Gomes et al., 2015; Yasur and Pathipati, 2015; Ferdous and Nemmar, 2020; Malhotra et al., 2020). The amount of available energy can vary under certain conditions and in a given life stage of an organism. It is a direct consequence of the accessible food resources and the ability to use them, that is, in animals, the efficiency of the digestive system and its supporting microorganisms (Karasov and Douglas, 2013; Celi et al., 2017). Hormesis may not occur if an organism does not have access to sufficient nutrients. In 2017, Wang et al. described an antibiotic-induced hormetic effect in *E. coli* only for bacteria cultured in media rich in utilizable carbon. According to Liebig's law of the minimum, the described regularity can be, with certain limitations (Gorban et al., 2010), extended to other organisms and processes involved in hormesis.

During the experiment, the insects had unlimited access to food. Aside from the unlikely effect of limiting the availability of nutrients by nanoparticles, it can be assumed that the insects did not starve during the experiment. Nevertheless, the effect of hormesis disappeared in the last period of the experiment, when consumption and assimilation (days 6–10), and the activity of the vast majority of the gut enzymes (day 10 of exposure) returned to the values typical for the control group (Figs. 2, 3, 5B, 5D, 6B, and 6D). To explain this phenomenon, we should also consider other factors that can shape the final response of an organism. The relationship between hormesis and dose and exposure time is clear and well documented (Calabrese, 2005; Agathokleous and Calabrese, 2020). However, many factors, partly related to the time, such as the animal's age, health/condition, physiological state, other harmful substances and reproductive effort (general body burden), may also be significant. Previous studies and our results allow to propose a 'hormesis graph' showing hormetic response as a resultant effect of numerous internal and external variables (Fig. 7). It should be noted that exposure time is only partially related to age and depends on the overall length of the animal's life. Aging organisms have a lower chance of surviving adverse conditions, weaker defense/repair mechanisms, and fewer energy reserves (Augustyniak et al., 2009a; Augustyniak et al., 2009b; Augustyniak et al., 2011). For *A. domesticus*, the period of 10 days of imago life was approximately ⅓ of the entire imago stage. Thus, the effect of hormesis can diminish

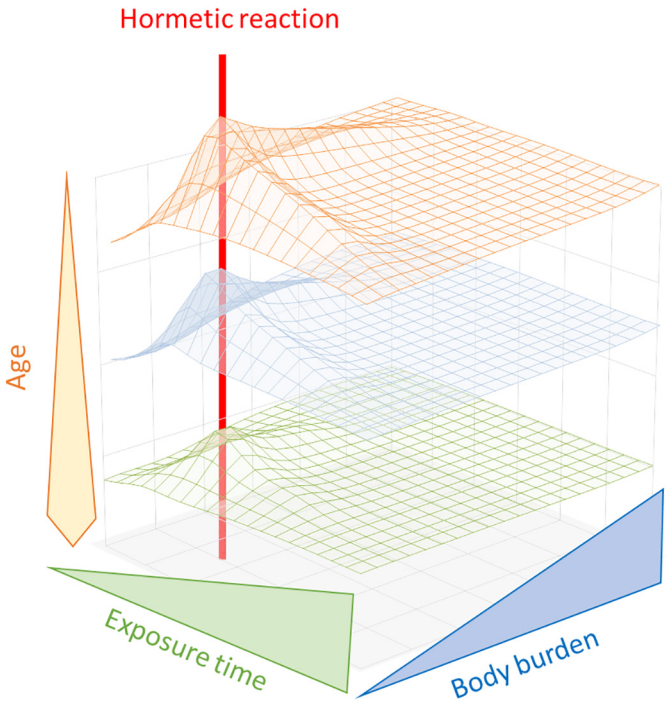

Fig. 7. Graphical presentation of hormetic response that depends on some internal and external variables. Explanations to the figure in the text.

with the exposure time and progressive aging of the animal (Fig. 7). In addition, the accumulation of other stress factors (toxins, metabolites, and degraded molecules) shapes the overall "body burden" and can influence the hormetic response. In *A. domesticus*, there were no additional stress factors. However, essential for the insects' overall "body burden" is the convergence of the disappearance of the hormetic effect on days 6–10 with the physiological need to produce eggs and oviposition. This phenomenon is cyclical and begins around this time (Destephano et al., 1982; Murtaugh and Denlinger, 1985).

Compared to the influence of individual nanoparticles on the tested parameters, it can be concluded that the GO-AgNP composite did not cause a drastically greater reaction than GO or AgNPs separately. The idea of an active nanoparticle surface, which can interact with the biomaterial, can help understand the obtained results. The large surface area of nanoparticles is responsible for their high reactivity (Jeevanandam et al., 2018). In the GO-AgNP composite, bonds between GO and AgNPs were formed, deactivating a part of the NP surface. Moreover, larger agglomerates were observed. This situation can lead to a reduction in the overall active area. Therefore, the GO-AgNP effect is weaker than it can be assumed, taking into account the simplified calculation, that is, the sum of the GO and AgNPs areas, used in this experiment. Thus, the composite may display lower activity with biological structures than its separate components. Further advanced material measurements and functional surface calculations are necessary to address this issue.

In the group treated with the composite, we found an increased amount of water in the feces (during the 3–6 days and 6–10 d intervals) and in the body of *A. domesticus* compared to the other experimental groups (Figs. 4C and D). Thus, the insects retained and possibly drank more water, which might have resulted from the greater water-holding capacity of the composite. Such effects were previously observed in plants grown on substrates with the addition of carbon nanoparticles (Khodakovskaya et al., 2012; Zhang et al., 2015; He et al., 2018; Park et al., 2020). To the best of our knowledge, this is the first observation of the effect of NPs on the water balance in insects. The effects of long-term exposure to the composite and greater water uptake importance will be investigated in the future.

5. Conclusions

This study confirms the hypothesis that used nanoparticles can change gut functions and deregulate the energy budget in the early stage of exposure. Even low amounts of dietary NPs can provoke an early physiological response that is stimulatory rather than inhibitory and resembles hormesis. Therefore, despite the different mode of deleterious action of nanoparticles, the exposed organisms seemed to mobilize a "standard" defense system that manifests itself by the alarm stage of the stress response to environmental threats. The age of the animal and the overall burden of the organism caused by reproduction or other factors may also influence the hormetic effect. However, this aspect should be examined in more advanced experiments, including a more extended period of life, critical stages in developing a given species, and, possibly, varus mixtures of xenobiotics.

CRediT authorship contribution statement

Reyhaneh Seyed Alian: Methodology, Investigation, Formal analysis, Writing – original draft, Visualization. **Marta Dziewięcka:** Methodology, Investigation, Validation, Formal analysis, Writing – original draft, Visualization. **Andrzej Kędziorski:** Methodology, Investigation, Validation, Formal analysis, Writing–original draft, Visualization. **Łukasz Majchrzycki:** Methodology, Validation, Investigation, Writing – review & editing. **Maria Augustyniak:** Conceptualization, Formal analysis, Writing–original draft, Visualization, Supervision, Project administration, Funding acquisition.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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References

Agathokleous, E., Calabrese, E.J., 2020. A global environmental health perspective and optimization of stress. Sci. Total Environ. 704, 135263. https://doi.org/10.1016/j.scitotenv.2019.135263.

Agathokleous, E., Feng, Z.Z., Iavicoli, I., Calabrese, E.J., 2019. The two faces of nanomaterials: A quantification of hormesis in algae and plants. Environ. Int. 131, 105044. https://doi.org/10.1016/j.envint.2019.105044.

Augustyniak, M., Babczyńska, A., Augustyniak, M., 2009a. Does the grasshopper Chorthippus brunneus adapt to metal polluted habitats? A study of glutathione-dependent enzymes in grasshopper nymphs. Insect Sci. 16, 33–42. https://doi.org/10.1111/j.1744-7917.2009.00251.x.

Augustyniak, M., Tarnawska, M., Babczyńska, A., Augustyniak, M., 2009b. Hsp70 level in progeny of aging grasshoppers from variously polluted habitats and additionally exposed to zinc during diapause. J. Insect Physiol. 55, 735–741. https://doi.org/10.1016/j.jinsphys.2009.04.009.

Augustyniak, M., Babczyńska, A., Augustyniak, M., 2011. Oxidative stress in newly-hatched Chorthippus brunneus—the effects of zinc treatment during diapause, depending on the female's age and its origins. Comp. Biochem. Physiol. C 154, 172–179. https://doi.org/10.1016/j.cbpc.2011.05.004.

Bao, Q., Zhang, D., Qi, P., 2011. Synthesis and characterization of silver nanoparticle and graphene oxide nanosheet composites as a bactericidal agent for water disinfection. J. Colloid Interface Sci. 360 (2), 463–470. https://doi.org/10.1016/j.jcis.2011.05.009.

Bhunia, S.K., Jana, N.R., 2014. Reduced Graphene oxide-silver nanoparticle composite as visible light Photocatalyst for degradation of colorless endocrine disruptors. ACS Appl. Mater. 6, 20085–20092. https://doi.org/10.1021/am505677x.

Boetius, A., Felbeck, H., 1995. Digestive enzymes in marine invertebrates from hydrothermal vents and other reducing environments. Mar. Biol. 122, 105–113. https://doi.org/10.1007/BF00349283.

Calabrese, E.J., 2005. Paradigm lost, paradigm found: the re-emergence of hormesis as a fundamental dose response model in the toxicological sciences. Environ. Pollut. 138, 378–411. https://doi.org/10.1016/j.envpol.2004.10.001.

Celi, P., Cowieson, A.J., Fru-Nji, R.F., Steinert, R.E., Kluenter, A.-M., Verlhac, V., 2017. Gastrointestinal functionality in animal nutrition and health: new opportunities for sustainable animal production. Anim. Feed Sci. Technol. 234, 88–100. https://doi.org/10.1016/j.anifeedsci.2017.09.012.

Cobos, M., De-La-Pinta, I., Quindós, G., Fernández, M.J., Fernández, M.D., 2020. Graphene oxide-silver nanoparticle nanohybrids: synthesis, characterization, and antimicrobial properties. Nanomaterials. 10 (2), 376. https://doi.org/10.3390/nano10020376.

Cohen, E., 2005. Pesticide-mediated homeostatic modulation in arthropods. Pestic. Biochem. Physiol. 85 (1), 21–27. https://doi.org/10.1016/j.pestbp.2005.09.002.

Collin, R., Starr, M.J., 2013. Comparative ontogenetic changes in enzyme activity during embryonic development of Calyptraeid gastropods. Biol. Bull. 225, 8–17. https://www.jstor.org/stable/23595252.

Cutler, G.Ch, 2013. Insects, insecticides and hormesis: evidence and considerations for study. Dose-Response. 11, 154–177. https://doi.org/10.2203/dose-response.12-008.Cutler.

de Medeiros, A.M.Z., Khan, L., da Silva G, U., Ospina C, H., Alves O, A., de Castro L, L., Martinez D, V., T, S., 2021. Graphene oxide-silver nanoparticle hybrid material: an integrated nanosafety study in zebrafish embryos. Ecotoxicol. Environ. Saf. 209, 111776. https://doi.org/10.1016/j.ecoenv.2020.111776.

de Saravia, S.G.G., Rastelli, S.E., Angulo-Pineda, C., Palza, H., Viera, M.R., 2020. Anti-adhesion and antibacterial activity of silver nanoparticles and graphene oxide-silver nanoparticle composites. Rev. Mater. 25 (2). https://doi.org/10.1590/S1517707620200002.1071.

Destephano, D.B., Brady, U.E., Farr, C.A., 1982. Factors influencing oviposition behavior in the cricket, Acheta domesticus. J. Econ. Entomol. 75, 111–114. https://doi.org/10.1093/aesa/75.2.111.

Dideikin, A.T., Vul, A.Y., 2019. Graphene oxide and derivatives: the place in graphene family. Front. Phys. 6, 149. https://doi.org/10.3389/fphy.2018.00149.

Dinh, D.A., Hui, K.S., Hui, K.N., Cho, Y.R., Zhou, W., Hong, X., Chun, H.H., 2014. Green synthesis of high conductivity silver nanoparticle-reduced graphene oxide composite films. Appl. Surf. Sci. 298, 62–67. https://doi.org/10.1016/j.apsusc.2014.01.101.

Doherty-Weason, D., Oyarzun, F.X., Vera, L., Bascur, M., Guzmán, F., Silva, F., Urzúa, Á., Brante, A., 2019. Role of the larval feeding morphology and digestive enzyme activity in the early development of the polychaete Boccardia wellingtonensis. PeerJ. Aquatic Biology. 7, e6151. https://doi.org/10.7717/peerj.6151.

Dziewięcka, M., Karpeta-Kaczmarek, J., Augustyniak, M., Majchrzycki, Ł., Augustyniak-Jabłokow, M.A., 2015. Evaluation of in vivo graphene oxide toxicity for Acheta domesticus in relation to nanomaterial purity and time passed from the exposure. J. Hazard. Mater. 305, 30–40. https://doi.org/10.1016/j.jhazmat.2015.11.021.

Dziewięcka, M., Karpeta-Kaczmarek, J., Augustyniak, M., Rost-Roszkowska, M., 2017. Short-term in vivo exposure to graphene oxide can cause damage to the gut and estis. J. Hazard. Mater. 328, 80–89. https://doi.org/10.1016/j.jhazmat.2017.01.012.

Dziewięcka, M., Witas, P., Karpeta-Kaczmarek, J., Kwaśniewska, J., Flasz, B., Balin, K., Augustyniak, M., 2018. Reduced fecundity and cellular changes in Acheta domesticus after multigenerational exposure to graphene oxide nanoparticles in food. Sci. Total Environ. 635, 947–955. https://doi.org/10.1016/j.scitotenv.2018.04.207.

Dziewięcka, M., Flasz, B., Rost-Roszkowska, M., Kedziorski, A., Kochanowicz, A., Augustyniak, M., 2020. Graphene oxide as a new anthropogenic stress factor – multigenerational study at the molecular, cellular, individual and population level of Acheta domesticus. J. Hazard. Mater. 396, 122775. https://doi.org/10.1016/j.jhazmat.2020.122775.

Ferdous, Z., Nemmar, A., 2020. Health impact of silver nanoparticles: a review of the biodistribution and toxicity following various routes of exposure. Int. J. Mol. Sci. 21, 2375. https://doi.org/10.3390/ijms21072375.

Flasz, B., Dziewięcka, M., Kędziorski, A., Tarnawska, M., Augustyniak, M., 2020. Vitellogenin expression, DNA damage, health status of cells and catalase activity in Acheta domesticus selected according to their longevity after graphene oxide treatment. Sci. Total Environ. 737, 140274. https://doi.org/10.1016/j.scitotenv.2020.140274.

Gao, F., Hui Liu, X., Ge, F., 2007. Energy budgets of the Chinese green lacewing (Neuroptera: Chrysopidae) and its potential for biological control of the cotton aphid (Homoptera: Aphididae). Insect Sci. 14 (6). https://doi.org/10.1111/j.1744-7917.2007.00178.x.

Gomes, I.L., Hansen, D., Scott-Fordsmand, J., Amorim, M.J.B., 2015. Effects of silver nanoparticles to soil invertebrates: oxidative stress biomarkers in Eisenia fetida. Environ. Pollut. 199, 49–55. https://doi.org/10.1016/j.envpol.2015.01.012.

Gorban, A., Pokidysheva, L., Smirnova, E., Tyukina, T., 2010. Law of the minimum paradoxes. Bull. Math. Biol. 73 (9), 2013–2044. https://doi.org/10.1007/s11538-010-9597-1.

Guedes, R.N.C., Magalhães, L.C., Cosme, L.V., 2009. Stimulatory sublethal response of a generalist predator to Permethrin: Hormesis, Hormoligosis, or homeostatic regulation? J. Econ. Entomol. 102 (1), 170–176. https://doi.org/10.1603/029.102.0124.

Guo, X., Mei, N., 2014. Assessment of the toxic potential of graphene family nanomaterials. J. Food. Drug. 22, 105–115. https://doi.org/10.1016/j.jfda.2014.01.009.

Gurunathan, S., Han, J.W., Park, J.H., Kim, E., Y-J, Choi, D-N, Kwon, J-H, Kim, 2015. Reduced graphene oxide–silver nanoparticle nanocomposite: a potential anticancer nanotherapy. Int. J. Nanomedicine 10, 6257–6276.

He, Y., Hu, R., Zhong, Y., Zhao, X., Chen, Q., Zhu, H., 2018. Graphene oxide as a water transporter promoting germination of plants in soil. Nano Res. 11, 1928–1937. https://doi.org/10.1007/s12274-017-1810-1.

Hoffmann, G.R., Stempsey, W.E., 2008. The hormesis concept and risk assessment: are there unique ethical and policy considerations? Hum. Exp. Toxicol. 27 (8), 613–620. https://doi.org/10.1177/0960327108098487.

Horch, H.W., Mito, T., Popadi, A., Ohuchi, H., Noji, S., 2017. The Cricket as a Model Organism Development, Regeneration, and Behavior. Springer Japan, Tokyo https://doi.org/10.1007/978-4-431-56478-2.

Iavicoli, I., Calabrese, E.J., Nascarella, M.A., 2010. Exposure to nanoparticles and hormesis. Dose-Response 8, 501–517. https://doi.org/10.2203/dose-response.10-016.Iavicoli.

Iavicoli, I., Leso, V., Fontana, L., Calabrese, E.J., 2018. Nanoparticle exposure and Hormetic dose–responses: an update. Int. J. Mol. Sci. 19, 805. https://doi.org/10.3390/ijms19030805.

Inshakova, E., Inshakov, O., 2017. World Market for Nanomaterials: Structure and Trends. 129. MATEC Web of Conferences, p. 02013. https://doi.org/10.1051/matecconf/201712902013.

Jeevanandam, J., Barhoum, A., Chan, Y.S., Dufresne, A., Danquah, M.K., 2018. Review on nanoparticles and nanostructured materials: history, sources, toxicity and regulations. Beilstein J. Nanotechnol. 3, 1050–1074. https://doi.org/10.3762/bjnano.9.98.

Karasov, W.H., Douglas, A.E., 2013. Comparative digestive physiology. Compr. Physiol. 3 (2), 741–783. https://doi.org/10.1002/cphy.c110054.

Karpeta-Kaczmarek, J., Augustyniak, M., Rost-Roszkowska, M., 2016a. Ultrastructure of the gut epithelium in Acheta domesticus after long- term exposure to nanodiamonds supplied with food. Arthropod. Struct. Dev. 45, 253–264. https://doi.org/10.1016/j.asd.2016.02.002.

Karpeta-Kaczmarek, J., Dziewięcka, M., Augustyniak, M., Rost-Roszkowska, M., 2016b. Effects of short-term exposure of Acheta domesticus to nanodiamonds in food: DNA damage but no histological alteration in tissues. Carbon 110, 458–468. https://doi.org/10.1016/j.carbon.2016.09.053.

Karpeta-Kaczmarek, J., Dziewięcka, M., Augustyniak, M., Rost-Roszkowska, M., Pawlyta, M., 2016c. Oxidative stress and genotoxic effects of diamond nanoparticles. Environ. Res. 148, 264–272. https://doi.org/10.1016/j.envres.2016.03.033.

Karpeta-Kaczmarek, J., Kędziorski, A., Augustyniak-Jabłokow, M.A., Dziewięcka, M., Augustyniak, M., 2018. Chronic toxicity of nanodiamonds can disturb development and reproduction of Acheta domesticus L. Environ. Res. 166. https://doi.org/10.1016/j.envres.2018.05.027.

Katz, L.M., Dewan, K., Bronaugh, R.L., 2015. Nanotechnology in cosmetics. Food Chem. Toxicol. 85, 127–137. https://doi.org/10.1016/j.fct.2015.06.020.

Kaufman, M.G., Klug, M.J., Merritt, R.W., 1989. Growth and food utilization parameters of germ-free house crickets *Acheta domesticus*. J. Insect Physiol. 35 (12), 957–967. https://doi.org/10.1016/0022-1910(89)90019-X.

Kavinkumar, T., Krishnamoorthy, V., Ravikumar, V., Manivannan, S., 2017. Anticancer activity of graphene oxide-reduced graphene oxide-silver nanoparticle composites. J. Colloid Interface Sci. 505, 1125–1133. https://doi.org/10.1016/j.jcis.2017.07.002.

Khodakovskaya, M.V., Silva, K.D., Biris, A.S., Dervishi, E., Villagarcia, H., 2012. Carbon nanotubes induce growth enhancement of tobacco cells. ACS Nano 6 (3), 2128–2135. https://doi.org/10.1021/nn204643g.

Li, I., Tang, M., Xue, Y., 2018. Review of the effects of silver nanoparticle exposure on gut bacteria. J. Appl. Toxicol. 39 (1), 27–37. https://doi.org/10.1002/jat.3729.

Luckey, T.D., 1968. Insecticide hormoligosis. J. Econ. Entomol. 61, 7–12. https://doi.org/10.1093/jee/61.1.7.

Malakar, A.S., Kanel, R., Ray, C., Snow, D.D., Nadagouda, M.N., 2021. Nanomaterials in the environment, human exposure pathway, and health effects: a review. Sci. Total Environ. 759, 143470. https://doi.org/10.1016/j.scitotenv.2020.143470.

Malhotra, N., Villaflores, O.B., Audira, G., Siregar, G., Lee, L.S., 2020. Toxicity studies on graphene-based nanomaterials in aquatic organisms: current understanding. Molecules 25, 3618. https://doi.org/10.3390/molecules25163618.

Mattson, M.P., 2008. Hormesis defined. Ageing Res Rev. 7 (1), 1–7. https://doi.org/10.1016/j.arr.2007.08.007.

McGillicuddy, E.M., Murray, I., Kavanagh, S., Morrison, L., Fogarty, A., Cormican, M., Dockery, P., Prendergast, M., Rowan, N., Morris, D., 2017. Silver nanoparticles in the environment: sources, detection and ecotoxicology. Sci. Total Environ. 575, 231–246. https://doi.org/10.1016/j.scitotenv.2016.10.041.

Murtaugh, M.P., Denlinger, D.L., 1985. Physiological regulation of long-term oviposition in the house cricket, *Acheta domesticus*. J. Insect Physiol. 8, 611–617. https://doi.org/10.1016/0022-1910(85)90059-9.

Park, S., Choi, K.S., Kim, S., Gwon, Y., Kim, J., 2020. Graphene oxide-assisted promotion of plant growth and stability. Nanomaterials. 10, 758. https://doi.org/10.3390/nano10040758.

Przemieniecki, S.W., Kosewska, A., Ciesielski, S., Kosewska, O., 2020. Changes in the gut microbiome and enzymatic profile of Tenebrio molitor larvae biodegrading cellulose, polyethylene and polystyrene waste. Environ. Pollut. 256, 113265. https://doi.org/10.1016/j.envpol (2019.113265.).

Pulit-Prociak, J., Banach, M., 2016. Silver nanoparticles – a material of the future? Open Chem. 14, 76–91. https://doi.org/10.1515/chem-2016-0005.

Rai, M., Yadav, A., Gade, A., 2009. Silver nanoparticles as a new generation of antimicrobials. Biotechnol. Adv. 27, 76–83. https://doi.org/10.1016/j.biotechadv.2008.09.002.

Schowalter, T., 2006. Insect Ecology. 2nd ed. Elsevier https://doi.org/10.1016/B978-012088772-9/50027-3.

Smith, A.T., LaChance, A.M., Zeng, S., Liu, B., Sun, L., 2019. Synthesis, properties, and applications of graphene oxide/reduced graphene oxide and their nanocomposites. Nano Materials Sci. 1, 31–47. https://doi.org/10.1016/j.nanoms.2019.02.004.

Szelei, J., Woodring, J., Goettel, M.S., Duke, G., Jousset, F.X., Liu, K.Y., Zadori, Z., Li, Y., Styer, E., Boucias, D.G., Kleespies, R.G., Bergoin, M., Tijssen, P., 2011. Susceptibility of NorthAmerican and European crickets to Acheta domesticus densovirus (AdDNV) and associated epizootics. J. Invertebr. Pathol. 106, 394–399. https://doi.org/10.1016/j.jip.2010.12.009.

Taglietti, A., Diaz Fernandez, Y.A., Amato, E., Cucca, L., Dacarro, G., Grisoli, P., Necchi, V., Dacarro, G., Grisoli, P., Necchi, V., Pallavicini, P., Pasotti, L., Patrini, M., 2012. Antibacterial activity of glutathione-coated silver nanoparticles against gram-positive and gram-negative bacteria. Langmuir. 28, 8140–8148. https://doi.org/10.1021/la3003838.

Tang, J., Chen, Q., Xu, L., Zhang, S., Feng, L., Cheng, L., Xu, H., Liu, Z., Peng, R., 2013. Graphene oxide−silver Nanocomposite as a highly effective antibacterial agent with species-specific mechanisms. ACS Appl. Mater. Interfaces 5, 3867–3874. https://doi.org/10.1021/am4005495.

Waldbauer, G.P., 1968. The consumption and utilization of food by insects. Adv. Insect Phys. 5, 229–288. https://doi.org/10.1016/S0065-2806(08)60230-1.

Wang, D., Lin, Z., Wang, T., Ding, X., Liu, Y., 2017. An analogous wood barrel theory to explain the occurrence of hormesis: A case study of sulfonamides and erythromycin on Escherichia coli growth. PLoS One 12 (7), e0181321. https://doi.org/10.1371/journal.pone.0181321.

Wei, L., Lu, J., Xu, H., Patel, A., Chen, Z.S., Chen, G., 2015. Silver nanoparticles: synthesis, properties, and therapeutic applications. Drug Discov. Today 20 (5), 595–601. https://doi.org/10.1016/j.drudis.2014.11.014.

Wu, C., Zhang, Y., Wu, X., Yang, Y., Zhou, X., Wu, H., 2012. Biological applications of graphene and graphene oxide. Nano Biomed. Eng. 4 (157–162), 157–162. https://doi.org/10.5101/nbe.v4i4.

Xue-Fei, S., Jing, Q., Peng-Fei, X., Bei-Bei, G., Chun-Miao, Y., Chao, S., Shu-Guang, W., 2015. Graphene oxide–silver nanoparticle membrane for biofouling control and water purification. Chem. Eng. J. 281, 53–59. https://doi.org/10.1016/j.cej.2015.06.059.

Yasur, J., Pathipati, U.R., 2015. Lepidopteran insect susceptibility to silver nanoparticles and measurement of changes in their growth, development and physiology. Chemosphere 124, 92–102. https://doi.org/10.1016/j.chemosphere.2014.11.029.

Yates, L.R., Daza, M., Sáiz, F., 2011. The energy budget of adult *Pseudopachymerina spinipes* (Er.) (Coleoptera: Bruchidae). Can. J. Zool. 67 (3), 721–726. https://doi.org/10.1139/z89-104.

Zhang, M., Gao, B., Chen, J., Li, Y., 2015. Effects of graphene on seed germination and seedling growth. J. Nanopart. Res. 17, 78. https://doi.org/10.1007/s11051-015-2885-9.

Zhu, Z., Su, M., Ma, L., Liu, D., Wang, Z., 2013. Preparation of graphene oxide–silver nanoparticle nanohybrids with highly antibacterial capability. Talanta 117, 449–455. https://doi.org/10.1016/j.talanta.2013.09.017.