Bull. Environ. Contam. Toxicol. (2002) 69:243–249

DOI: 10.1007/s00128-002-0053-2

Bulletin of Environmental Contamination and Toxicology

# Population Parameters of the Beetle *Pterostichus oblongopunctatus* F. from Metal Contaminated and Reference Areas

M. Łagisz, P. Kramarz, R. Laskowski, M. Tobor

Department of Ecotoxicology, Institute of Environmental Sciences, Jagiellonian University, Gronostajowa 3, 30-387 Kraków, Poland

Received: 21 August 2001/Accepted: 5 March 2002

One of the major problems in anthropogenicaly contaminated terrestrial ecosystems is the accumulation of metals in litter and surface soil layers due to the high affinity of metal ions to soil organic matter. Even at moderate inputs, metals can reach high concentrations after prolonged input with dust and rainfall (Benin et al. 1999; Kaminski and Landsberger 2000). The negative effect of metals in terrestrial ecosystems has been shown so far for litter decomposition processes and some soil-dwelling invertebrates (Hopkin and Martin 1985; Donker 1992; Cotrufo et al. 1995; Spurgeon and Hopkin 1996). However, our understanding of their influence on populations of soil and epigeic fauna is still far from complete. For example, it has been shown that some earthworm populations can tolerate heavy metal concentrations well above the concentration known to induce lethal effects in non-tolerant populations (Stürzenbaum et al. 1998). Results of a number of laboratory studies contradict some field observations in which viable populations of animals were found at soil contamination levels an order of magnitude higher than laboratory-established critical concentrations of metals in soil or food (e.g., Posthuma and Van Straalen 1993, Hopkin and Hames 1994). Such discrepancies suggest that, unfortunately, results derived from laboratory cultures cannot be easily translated to expected field effects and studies on field populations are indispensable.

Organisms inhabiting contaminated areas can be stressed by metal exposure for a prolonged time and are possibly subjected to selection for increased resistance to metals. This may result in physiological or behavioural adaptations to long-term sublethal metal exposure (Read et al. 1987; Donker and Bogert 1991; Posthuma and Van Straalen 1993). As an organism has to allocate energy to resist the toxicant by avoidance, exclusion, removal or complexation, this will inevitably decrease the amount of energy available for other processes such as growth or reproduction (Sibly and Calow 1989). Consequently, evolution of tolerance may lead to changes in a number of physiological and ecological characteristics: reproduction, transpiration and respiration, tolerance to additional environmental stressors such as food deprivation, insecticides, infections, and climatic stress (Holmstrup et al. 2000; Stone et al. 2001).

*Correspondence to:* M. Łagisz

**Table 1.** Concentrations of major pollutants in humus layer at the study sites (after Stone et al. 2001)

| **Site** | d [km] | Zn [mg kg$^{-1}$] | Cd [mg kg$^{-1}$] | Cu [mg kg$^{-1}$] | Pb [mg kg$^{-1}$] |
|---|---|---|---|---|---|
| 1 | 3.5 | 10454±2618.5 | 81.92±17.72 | 46.9±4.56 | 2635±120.4 |
| 2 | 2.5 | 5104±729.0 | 51.06±19.34 | 37.6±3.72 | 1832±215.0 |
| 3 | 3.9 | 1522±135.2 | 18.14±2.60 | 25.6±2.16 | 870±36.3 |
| 4 | 7.9 | 244±78.2 | 3.30±1.03 | 15.4±2.68 | 355±30.9 |
| 5 | 31.9 | 151±34.5 | 0.84±0.39 | 10.7±0.96 | 136±8.8 |

mean ± standard deviation, d – distance from the larger of the two smelters (Site1 is actually located between the two smelters, hence the contamination is higher that at Site2 despite somewhat larger distance from the larger smelter)

The ground beetles (Carabidae), as predators, represent the second trophic level, at least potentially exposed to elevated concentrations of toxicants. At the same time, they are known for low body metal concentrations even in highly contaminated areas (Hunter et al. 1987; Butovsky and Van Straalen 1995) due to effective excretion of assimilated metals (Janssen et al. 1991; Kramarz 1999). The detoxification process may require the expenditure of a significant amount of energy, as well as change in fitness-related life history traits. Knowledge of long-term pollutant effects on fitness in carabids has both scientific and applied value, as they are considered to be 'beneficial' insects. The aim of this study was to determine effects of prolonged metal contamination on population parameters, especially reproduction, in the forest-living ground beetle *Pterostichus oblongopunctatus* F. from five sites with different levels of metal pollution.

## MATERIALS AND METHODS

The carabid beetle *P. oblongopunctatus* is common to forests all over the Palearctic region. Individual size varies between 9 and 13 mm; females are 10% larger than males on the average. The sex ratio in captures is close to 1:1 most of the time and the mortality rate is equal for males and females (Brunsting 1981). *P. oblongopunctatus* is a spring breeder with summer larvae, and the adults hibernate. Reproduction occurs between the beginning of April and the end of July (Muller and Keschula, 1986).

Adult individuals of *P. oblongopunctatus* were collected with pitfall traps (Barber-type), during their reproductive period, from five sites along the soil pollution gradient. The most contaminated site (Site1) was located between two smelters, hence the soil contamination was much higher than at Site2 despite a somewhat larger distance (3.5 km) from the larger smelter. Distances from the larger smelter for the other sites were: 2.5 km (Site2), 3.9 km (Site3), 7.9 km (Site4), and 31.9 km (Site5). Metal concentrations in the organic soil layer ($O_{HF}$)

ranged from approximately 150 mg $kg^{-1}$ (Site5) to 10500 mg $kg^{-1}$ Zn (Site1). 136 to 2600 mg $kg^{-1}$ Pb, and 0.84 to 81.9 mg $kg^{-1}$ Cd (Tab.1) (Stone et al. 2001).

Twenty pairs of adults from Site1, 36 from Site2, 16 from Site3, 16 from Site4, and 24 from Site5 were used for culturing. All animals were kept in the laboratory 20±2°C, 16:8 LD), fed with uncontaminated food (Royal Canin, Kitten 34), and cultured according to the method described by Metge and Heimbah (1998) for the carnivorous staphylinid *Philonthus cognatus* Steph. For egg laying, each pair of beetles was kept in a separate plastic box (10 cm × 10 cm × 6 cm) with a net bottom (2 mm mesh) and a transparent plastic lid. The box was filled with a 2 cm layer of moistened, smooth surface, expanded clay granules and put into another box, which had a closed bottom. Two times a week, the eggs laid by the beetles were sifted out with water and collected with a fine brush. They were then placed individually on moist filter paper in a 24-well tissue culture plate for hatching. Plates were stored at 20 ± 2 °C in darkness and checked every day for hatched larvae. The hatched larvae were transferred separately into 30 ml plastic vials filled with moistened ground peat (pH 4.5-5). The larvae were cultured at 20±2°C and 16:8 LD, and were fed every 3 days until imagines emerged. Emerged adults were fed for the next 25 days, after which they were weighed and sexed.

Every individual from the $F_1$-generation had its own record card with the following information: individual symbol, egg collection date, emergence from pupae date, sex and imago body mass. For parent individuals (P-generation) collected in the field, the collection date, time of death, sex, body mass at the beginning of reproduction, number of eggs laid during first 6 weeks in a laboratory, proportion of hatched eggs, and proportion of imagines hatched from pupae were recorded. For statistical analysis logarithmic zinc concentration in soil (log(Zn)) was used as an index of pollution level. Each specimen was treated as an independent replicate. To assess relations between the level of metal contamination and individual life history parameters the General Linear Models procedure was used. For variables deviating significantly from the normal distribution (Kolmogorow-Smirnov test) the Kruskal-Wallis nonparametric test was used to distinguish differences between sites.

## RESULTS AND DISCUSSION

Field-collected females had significantly higher ($p<0.0001$) mean body mass than males, but no significant relation was found between average body mass of each sex and site contamination level ($p=0.966$). The proportion of egg-lying females was close to 0.8 at all sites. The number of eggs laid during the experiment was extremely variable: from 0 to 67 eggs per female. Pollution level had a significant effect on fecundity: number of eggs laid per female per week was negatively related to log(Zn), that is females from uncontaminated sites produced more eggs ($p=0.006$) (Fig.1).

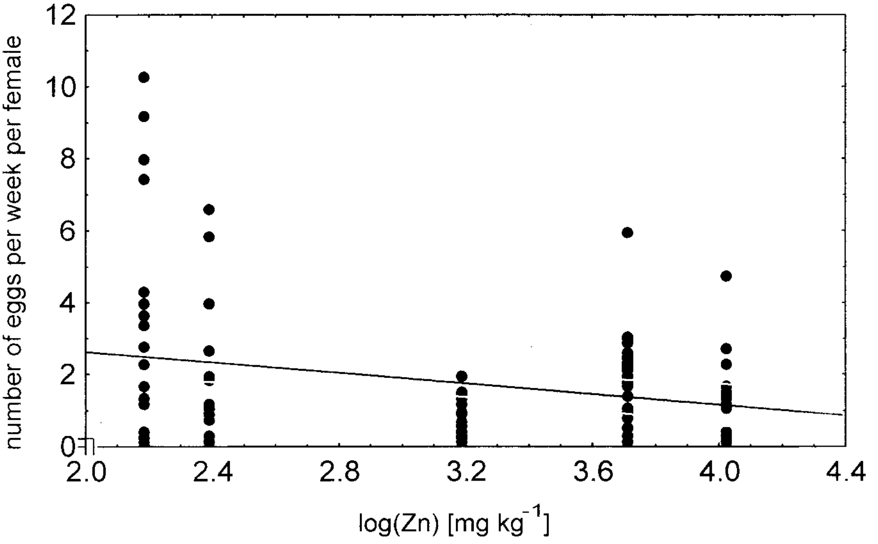


**Figure 1.** Correlation between pollution level expressed as log(zinc concentration) and total number of eggs per P-generation female
($y = -0.7307x + 4.0867$, $r = -0.2587$, $p = 0.0059$)

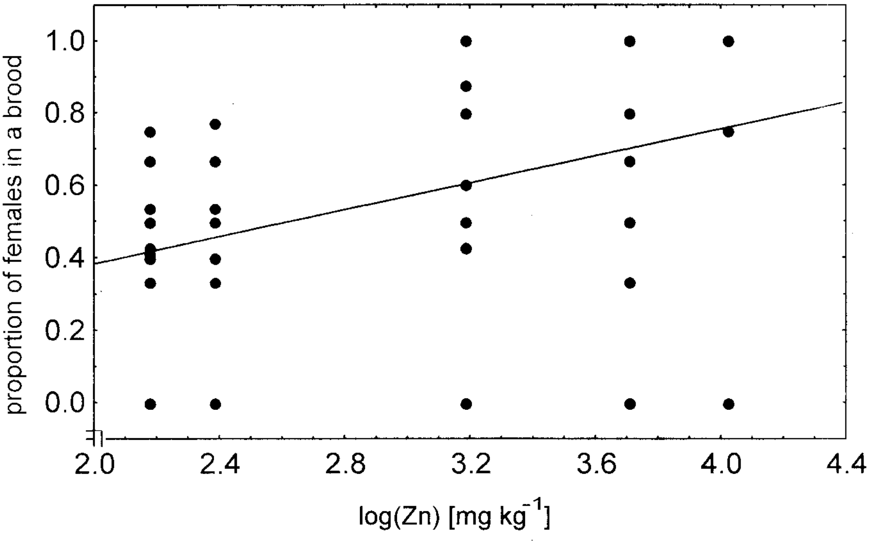


**Figure 2.** Correlation between pollution level expressed as log(zinc concentration) and proportion of females in the broods of P-generation females
($y = 0.1860x + 0.0106$, $r = 0.4380$, $p = 0.0179$)

**Table 2.** Population parameters of *P. oblongopunctatus* from five sites along a soil pollution gradient (mean ± standard deviation), N – number of P-generation pairs.

| Site | N | Number of eggs per female | Number of larvae per female | Larval survival rate | Number of adult offspring per female |
|---|---|---|---|---|---|
| 1 | 20 | 7.5±8.10 | 1.8±3.16 | 0.17±0.282 | 0.7±1.09 |
| 2 | 36 | 7.2±6.29 | 2.6±3.69 | 0.30±0.369 | 1.3±1.70 |
| 3 | 16 | 4.8±3.97 | 1.2±1.77 | 0.28±0.438 | 0.8±1.39 |
| 4 | 16 | 9.9±13.67 | 4.8±9.20 | 0.24±0.337 | 2.6±5.08 |
| 5 | 24 | 18.3±20.22 | 9.1±13.42 | 0.27±0.324 | 4.4±6.40 |

The hatching rate for eggs deviated significantly from the normal distribution ($p<0.05$) and did not differ between sites ($p=0.784$). For $F_1$-generation larvae that died before the imago stage was reached it was impossible to determine sex and the larval survival rate was calculated for both sexes together. There were no significant differences in larval survival rate between groups determined by the parents' habitat (Kruskal-Wallis test, $p=0.847$). However, for individuals that survived until the imago stage, the development time and body mass of males decreased with increasing log(Zn) ($p=0.033$ and $p=0.086$ respectively). No such relations were found for $F_1$ females ($p=0.401$ and $p=0.324$ respectively). At the same time, broods originating from the most contaminated sites had a significantly smaller proportion of males, while in the other broods the sex ratio was close to 1:1 ($p=0.018$) (Fig.2). This result deviated from the field situation in which the sex ratio was about 1:1 in all sites that P-generation beetles were collected from.

It has to be stressed that all larvae in the $F_1$ generation were fed standard uncontaminated food and were kept under identical conditions. Thus, any differences observed between $F_1$ individuals originating from different populations could be only due to either genetically fixed or maternal effects. The presence of some between-population differences in the first generation laboratory animals suggests that the differences between populations of *P. oblongopunctatus* from localities with different pollution levels could have a genetic basis. However, to determine if such a statement is valid further investigations on population parameters of $F_1$- and $F_2$-generation animals are necessary.

The overall reproductive success, measured as number of adult $F_1$ offspring per female, was significantly higher for P-females originating from uncontaminated sites than for females from polluted sites ($p=0.002$). The average number of offspring that survived to the imago stage was less than 1 per female from Site1 and more than 4 per female from Site5 (Tab.2).

The observed decrease in the reproductive rate in carabids originating form highly contaminated areas seems to support the hypothesis of a trade-off between efficient decontamination and productivity. Although carabids are able to control body levels of a number of metals almost perfectly (Janssen et al. 1991; Kramarz 1999) and direct toxicity should not be the case, they probably pay for this ability with a decreased amount of energy available for reproduction (Lindqvist and Block 2001). There are also indications that trade-offs may be due to pleiotropic effects of major genes. These costs are evident from reduced fitness of metal-adapted individuals in unpolluted environments and they might be due to interactions between tolerance against selected metal(s) and the metabolism of essential elements (Van Straalen and Hoffmann 2000). On the other hand, lower values of population parameters in contaminated localities may also be an indirect effect of differences in other field conditions, e.g. the expected scarcity of prey in the most polluted areas. Some studies indicate, however, that competition for food between carnivorous invertebrates may be even lower in polluted areas, where biodiversity often tends to decrease (Read et al. 1987). Other factors that can affect the life history parameters of the beetles are parasites and microclimatic differences between sites.

*Acknowledgments.* We thank the many individuals, who assisted us with collection of data in the field and in the laboratory, including Maria Niklińska, Maciej Maryański, Piotr Zygmunt and David Stone. Tom Mozdzer helped edit the manuscript. Financial support was provided by the National Committee for Scientific Research (Grant No 6 PO4F 043 18) and the Jagiellonian University.

## REFERENCES

Benin A, Sargent J, Dalton M, Roda S (1999) High concentrations of heavy metals in neighborhoods near ore smelters in northern Mexico. Environ Health Perspect 107:279-284

Brunsting AMH (1981) Distribution patterns, life cycle and phenology of *Pterostichus oblongopunctatus* F. (Col., Carabidae) and *Philonthus decorus* Grav. (Col., Staphylinidae). Netherlands J Zool 31:418-452

Butovsky RO, Van Straalen NM (1995) Copper and zinc contents in trophic chains of terrestial arthropods in the Moscow region. Pedobiologia 39:481-487

Cotrufo MF, Desanto AV, Alfani A, Bartoli G, Decristofaro A (1995) Effects of urban heavy metal pollution on organic matter decomposition in *Quercus ilex* L. woods. Environ Pollut 89:81-87

Donker MH (1992) Energy reserves and distribution of metals in populations of the isopod *Porcellio scaber* from metal contaminated sites. Func Ecol 6:445-454

Donker MH, Bogert CG (1991) Adaptations to cadmium in three populations of the isopod *Porcellio scaber*. Comp Biochem Physiol 100C:143-146

Holmstrup M, Bayley M, Sjursen H, Hojer R, Bossen S, Friis K (2000) Interactions between environmental pollution and cold tolerance of soil invertebrates: A neglected field of research. CryoLetters 21:309-314

Hopkin SP, Hames CAC (1994) Zinc, among a cocktail of metal pollutants, is responsible for the absence of the terrestrial isopod porcellio-scaber from the vicinity of a primary smelting works. Ecotoxicology 3: 68-78

Hopkin SP, Martin MH (1985) Transfer of heavy metals from leaf litter to terrestrial invertebrates. J Sci Food Agric 36:538-539
Hunter BA, Johnson. MS, Thompson. DJ (1987) Ecotoxicology of copper and cadmium in a contaminated grassland ecosystem. II. Invertebrates. J Appl Ecol 24:587-599
Janssen MPM, Bruins A, de Vries TH, Van Straalen NM (1991) Comparison of cadmium kinetics in four soil arthropod species. Arch Environ Contam Toxicol 20:305-312
Kaminski M, Landsberger S (2000) Heavy metals in urban soils of East St. Louis, IL, Part I: Total concentration of heavy metals in soils. J Air Waste Manag Assoc 50:1667-1679
Kramarz P (1999) Dynamics of accumulation and decontamination of cadmium and zinc in carnivorous invertebrates. 1. The ground beetle, *Poecilus cupreus* L. Bull Environ Contam Toxicol 63:531-537
Lindqvist L, Block M (2001) Metal pollution and fat accumulation in the carabid beetle *Pterostichus melanarius* (Coleoptera, Carabidae). Bull Environ Contam and Toxicol 66:184-188
Metge K, Heimbach U (1998) Tests on the Staphylinid *Philontus cognatus* Steph. 1832. In: Lökke H and Van Gestel CAM (ed) Handbook of Soil Invertebrate Toxicity Tests. John Wiley & Sons Ltd, p 157-179
Muller JK, Keschula A (1986) Biological significance of the seasonal distribution of activity of *P.oblongopunctatus* (F.) (Coleoptera, Carabidae). In: den Boer et al. (ed) Carabid beetles. Gustav Fisher, Stuttgart, New York
Posthuma L, Van Straalen NM (1993) Heavy-metal adapotation in terrestial invertebrates: a reviev of occurance, genetics, physiology and ecological conequences. Comp Biochem Physiol 106C:11-38
Read HJ, Wheater CP, Martin MH (1987) Aspects of the Ecology of Carabidae (Coleoptera) from woodlands polluted by heavy metals. Environ Pollut 48:61-76
Sibly RM, Calow P (1989) A life-cycle theory of responses to stress. Biol J Linn Soc 37:101-116
Spurgeon DJ, Hopkin SP (1996) The effects of metal contamination on earthworm populations around a smelting works: quantifying species effects. Appl Soil Ecol 4:147-160
Stone D, Jepson P, Kramarz P, Laskowski R (2001) Time to death response in carabid beetles exposed to multiple stressors along a gradient of heavy metal pollution. Environ Pollut 113:239-244
Stürzenbaum SR, Kille P, Morgan AJ (1998) Heavy metal-induced molecular responses in the earthworm *Lumbricus rubellus* genetic fingerprinting by directed differential display. Appl Soil Ecol 9:495-500
Van Straalen NM, Hoffmann AA (2000) Reviev of experimantal evidence for physiological costs of tolerance to toxicants. In: Kammenga J, Laskowski R (eds.) Demography in Ecotoxicology. p 147-161