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Estimation of required sampling effort for monitoring the possible effects of transgenic crops on butterflies: Lessons from long-term monitoring schemes in Switzerland

Andreas Lang[a,*], Christoph Bühler[b]

[a] University of Basel, Institute of Environmental Geosciences, Bernoullistrasse 30, CH-4056 Basel, Switzerland
[b] Hintermann & Weber AG, Austrasse 2a, CH-4153 Reinach, Switzerland

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ABSTRACT

Butterflies (Lepidoptera) have been suggested for environmental monitoring of genetically modified organisms (GMO) due to their suitability as ecological indicators, and because of the possible adverse impact of the cultivation of current transgenic crops. A critical point is the sampling effort to be invested in such a monitoring. Here, we estimated the required sample size necessary to monitor potential effects of genetically modified crops on butterflies (Lepidoptera).

We used data from two Swiss long-term butterfly monitoring surveys applying the common transect count method. The two monitoring surveys differed in several basic aspects such as geographical area covered, landscape context and sampling intensity. We carried out prospective power analyses in order to estimate the required sample size to detect effects of differing magnitude on mean species number, total individual abundance, mobility classes of butterflies and selected individual species.

The required sample size decreased substantially when effect sizes above 10% were estimated. For example, a sample size of 79 transects would be sufficient to detect changes of 30% in total individual abundance for both survey types. Detecting effects on mean species number would need much less transects. Considerably more samples would be needed to analyze the abundance of single species. Several options are presented to increase statistical power or reduce required sample size, respectively. Also, we recommend to pool species to different mobility classes, and/or analyze patch occupancy of species instead of their individual abundance.

The transect count approach is a suitable method for butterfly monitoring, both on a local as well as on a landscape scale. Consequently, both types of Swiss butterfly monitoring schemes are basically suitable for GMO monitoring. If transects are short and restricted to intensely used landscape, even non-professional field workers may yield data sufficient for effective monitoring, which might be relevant with respect to involved costs.

1. Introduction

Butterflies and day-active moths (Lepidoptera) are particularly suitable for environmental monitoring purposes as the species can be identified and monitored straightforwardly, are well-studied and well-documented in terms of faunistics, ecology and Red Book status, and they are significantly related to diverse conditions in ecosystems at different spatial and temporal levels (e.g., Oostermeijer and van Swaay, 1998; Pollard et al., 1995; Roth et al., 2008; Schmeller et al., 2009; Settele et al., 1999; Thomas, 2005; Van Dyck et al., 2009; but see Fleishman and Murphy, 2009). In addition, there exists a standardized methodology to monitor butterflies, the transect count method, which is described in detail and is widely accepted (e.g., Pollard and Yates, 1993; VDI, 2010). Hence, butterflies and day-active moths are frequently used in environmental monitoring across Europe (Van Swaay et al., 2008), e.g. recently as one of the two indicator taxa to assess biodiversity change in Europe (EEA, 2007, 2010).

Butterflies have been suggested for environmental GMO monitoring due to their suitability as ecological indicators, due to the possible adverse impact of the cultivation of transgenic crops on Lepidoptera, and because they are generally valued as relevant protection goals (Graef et al., 2005; Haughton et al., 2003; Lang and Vojtech, 2006; Lang and Otto, 2010; VDI, 2010). However, the possible design, range of use, operating conditions and practicability of a specific GMO butterfly monitoring are still under debate. A critical point is the sample size to be taken, i.e. the number of necessary

* Corresponding author. Fax: +41 061 267 0479.
E-mail addresses: andreas.lang@unibas.ch (A. Lang), buehler@hintermannweber.ch (C. Bühler).

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Table 1
Comparison of the two Swiss butterfly monitoring schemes: the LANAG (long term monitoring of biodiversity in the landscape of Kanton Aargau) and the BDM (Biodiversity Monitoring Switzerland).

Parameter	LANAG	BDM
Geographic scale	Kanton Aargau	Whole Switzerland
Spatial scale (sampling unit)	Local (250 m transect)	Landscape (1 km^2 square)
Start	1998	2003
Inspections per transect	10 per year	7 per year
Transect numbers	517	520
Transect length	250 m	2.5 km
Transect width	10 m	10 m
Mean species richness	Lower	Higher
Personnel	Educated amateurs	Professional entomologists

data points to record a certain GMO effect, as this determines the operating and monetary cost. A prospective power analysis is generally recommended to estimate the required sample sizes and/or statistical power to detect effects of a given magnitude (Nakagawa and Foster, 2004; Perry et al., 2003; VDI, 2010). To our knowledge, only two reports have been published estimating a potential sampling effort for a butterfly GMO monitoring plan (Aviron et al., 2009; Lang, 2004), leading to inconsistent results regarding estimated sample sizes and conclusions about the feasibility of a GMO butterfly monitoring. While Lang's study (2004) lacks conclusions whether butterfly-monitoring is feasible in practice, Aviron et al. (2009) consider case-specific butterfly monitoring to be too laborious and costly for practical use. However, the data analyzed by Lang (2004) and Aviron et al. (2009) were both not based on the standardized transect count method, the most accepted and applied methodology to monitor butterflies and day-active moths. In addition, the data used in both studies stem from a limited number of field-seasons. This may affect estimations of trend and/or variances because butterflies are short-lived insects known to show temporary fluctuation of abundances, often due to weather-conditions of a particular or previous year, or inherent population dynamics (e.g., Roy et al., 2001; Wilson and Roy, 2009). Hence, it remains uncertain if the results of these two studies apply to common butterfly monitoring schemes unequivocally.

Required sample size and proven effectiveness of recording schemes are preconditions for installing a butterfly monitoring programme to observe potential adverse GMO-induced effects. Here, we present results from two comprehensive long-term Swiss monitoring schemes for butterflies and day-active moths to address the question of sampling effort for detecting different effect sizes. Both schemes apply the common transect count method but differ in transect length, sampling intensity and various other aspects. Specifically, we (i) conducted a prospective power analysis based on the variability of the actual count data of the two monitoring programmes, (ii) deduced required sample size to record given GMO effects, and (iii) discuss the possibility of using the differing methods of these two routine monitoring programmes for the purpose of GMO monitoring.

2. Material and methods

2.1. Data sources

For the analyses of the present study, we used datasets of two different Swiss routine monitoring schemes (Table 1). The "Biodiversitätsmonitoring Schweiz" (BDM, "Biodiversity Monitoring Switzerland", www.biodiversitymonitoring.ch, last access on 1 December 2010) covers the whole country's area of Switzerland and the "Langfristbeobachtung der Artenvielfalt in der Normallandschaft des Kanton Aargau" (LANAG, "Long term monitoring of biodiversity in the "normal" landscape of Kanton Aargau", www.ag.ch/alg/de/pub/natur_landschaft/erfolgskontrolle/lanag.php, last access on 1 December 2010) is restricted to only one county (canton) in the north of Switzerland.

2.2. Sampling concepts

Both schemes collect data for butterflies and day-active moths from regularly distributed sites on a grid enabling stratified sampling to e.g. regional topography. In the LANAG Scheme 517 sites (butterfly transects) were established and for the BDM there are 520 sites (butterfly transects). Both schemes use a sampling approach shifted in time where a regular subset of one-fifth of the total of sites is sampled per year. Thus, it takes five years to complete the whole sample, and on every sixth year the sampling cycle starts again and the plots (transects) are re-assessed. Because the sampling locations remain stationary, paired measures for all sampling units will be available after 10 years. So, for each sampling unit there are exactly 5 years between census 1 and census 2. Note that, as the BDM has started in 2003, only a fraction of these transects have been monitored twice and could be used for the analysis of matched pairs (see below).

Apart from the above similarities, the LANAG and the BDM scheme differ in several points, in particular in the spatial scale covered (Table 1). LANAG focuses on a more local or habitat scale using transects of total length of 250 m. In contrast, BDM uses transects of 2.5 km that are aimed to reflect the butterfly community of a whole landscape section. Other important features that distinguish the two monitoring schemes are the number of visits per season and the education of the personnel involved in fieldwork (Table 1). Collection of field data for butterflies started in 1998 for LANAG, and in 2003 for BDM.

2.3. Data collection: LANAG

For the LANAG scheme 10 inspections were made per 250 m transect each year between April and September, during standardized favorable weather conditions (cf. Roth et al., 2008). The inspections were performed mostly by non-biologists (amateurs) prepared by a special training and supervised by professionals. At each inspection, the transect routes were walked once in both directions recording all of the observed butterflies within a 5-m band of the transect line. If possible, all butterfly individuals (including Zygaenidae) within the transect area were identified to the species level except for the *Pieris* genus where all species are aggregated within one taxon *Pieris* sp. agg. The family Hesperiidae was completely ignored in the field because the amateurs doing the field-work were known to vary considerably in their ability to detect the individuals of this family. All individuals of all species encountered within the transect area were noted and counted, the total for all the inspections giving an estimate of species abundance for this transect. For each transect, land use information for the transect route was mapped according to the CORINE Land Cover system.

2.4. Data collection: BDM

To obtain butterfly data for the BDM samples, seven inspections of a 2.5 km transect were conducted between April and September. In a preliminary study (unpublished), seven inspections were assessed to yield an adequate relationship of number of visits and proportion of the local species pool observed. Experienced and specially trained professional observers walked the transects in both directions during standardized and favorable weather conditions (cf. Pearman and Weber, 2007). All observed day-flying butterfly species (including Hesperiidae and Zygaenidae) $\leq$ 5 m