

Non-target Lepidoptera on Southern Vancouver Island:

**Field assessment at four years after the
1999 gypsy moth eradication program**

2003 Final Report

Timothy J. Boulton

18 July, 2003

Table of Contents

Acknowledgements	3
Project Summary.....	4
Research Paper on the 2003 field season	
Introduction	7
Methods	9
Results	11
Discussion.....	23
Analysis of the non-target Lepidoptera monitoring program (1999-2004)	
Initial impacts of <i>Btk</i> on non-target Lepidoptera	28
Recovery of non-target Lepidoptera	31
Beneficial side-effects of <i>Btk</i> (i.e. reductions of pest species)	37
Recommendations.....	38
References.....	39
Appendices	
A. Table of species composition for 1999	42
B. Table of species composition for 2000	43
C. Table of species composition for 2003	44

Acknowledgements

I wish to extend my sincere gratitude to the following individuals and institutions for their support and assistance during the course of this study: The province of British Columbia provided funding for this project and Peter Hall (Forest Practices Branch, BC Ministry of Forests) provided practical support; Robert Duncan (Canadian Forest Service) provided considerable taxonomic expertise and without his efforts many species could not have been identified; Darya Lis assisted in the field, Neil Borecky provided cartographic expertise and Martin Voordouw helped analyze the data; 'The Friends of Government House', and the municipalities of Esquimalt, Oak Bay, Saanich, and Victoria authorized access to parkland for research purposes.

Project Summary

The present study was initiated to measure the immediate impacts (1999-2000) of the eradication program on non-target Lepidoptera and to estimate the level of recovery that had occurred by the spring of 2003, four years after the *Btk* applications. The 2003 data are the focus of this report but the major conclusions drawn from the first two years of the study are also summarized and discussed in the light of the current data. The 1999 and 2000 reports may be obtained from the British Columbia Ministry of Forests.

1999: Year of the *Btk* applications

During 1999, year one of the study, 16,218 lepidopteran larvae were collected from 588 Garry oak branches from 26 April – 6 July. A total of 10,050 lepidopteran larvae were collected from 1,200 snowberry branches from 25 March – 15 June. Most (9/14) of the abundant Lepidoptera species on Garry oak and common snowberry were significantly ($p=0.05$) reduced in number within 5 weeks of the first *Btk* application. Most of the lepidopteran species were collected very infrequently because of sparse distributions, and were therefore pooled together for statistical analysis. As a group, these less common species were significantly reduced by *Btk*. Lepidoptera diversity was reduced in sprayed areas, compared to check areas, on Garry oak and common snowberry, according to the Margalef and Simpson diversity index, respectively. Species richness also was reduced on both plant species as a result of the pesticide.

2000: One year after the *Btk* applications

During 2000, year two of the study, immature Lepidoptera were sampled in 22 of the original 28 Garry oak woodlands from 25 April to 23 June. Immature Lepidoptera were collected from snowberry at 20 of the original 24 snowberry sites from 27 March to 1 June. A total of 7,132 lepidopteran larvae were collected from 594 Garry oak branches, and 4,060 lepidopteran larvae were collected from 1,000 snowberry branches. Each of the ten most abundant species on Garry oak and common snowberry was significantly less abundant in the treatment sites than the check sites. As a group, the less common species were also significantly reduced by *Btk*. Lepidoptera diversity was reduced in sprayed areas, compared to check areas, on Garry oak and common snowberry, according to the Margalef and Simpson diversity index, respectively. Species richness also was reduced on both plant species as a result of the pesticide.

2003: Four years after the *Btk* applications

During 2003, four years after the *Btk* applications, 6,790 lepidopteran larvae were collected from 200 Garry oak branches from 10 May – 3 June. Snowberry was not sampled in 2003. Sampling on Garry oak was performed using the same protocol that was used in the previous years of this study except that the number of sites, sample dates and Garry oak trees sampled were reduced in 2003. Among the six most abundant species in the 2003 samples, only one (*O. brumata*) was significantly more abundant in the check plots than the treatment plots; and only one species (*E. emarginana*) was significantly more abundant in the treatment plots than the check plots. Among the remaining 31 species collected in 2003, only two (*Y. cervella* and *E. vancouverensis*) were significantly more abundant in the check plots than the treatment plots, and none were significantly more abundant in the check plots. As a group, the uncommon species (i.e. those collected fewer than 60 times) were not significantly different between the treatment and check plots, however species richness per branch remained significantly lower in the check plots.

1999-2003: Main conclusions

The results from year one of the study indicated that the *Btk* applications significantly affected Lepidoptera populations in the short-term. The results in the second year showed that the impact of *Btk* on non-target Lepidoptera was more severe at 12-13 months post-spray than 1-2 months post-spray, and that by 2000 the affected species had not yet begun the recovery process. At 12-13 months post-spray, four additional species were shown to be reduced by the 1999 *Btk* applications, indicating that some effects went undetected in 1999. The data in 2000 also indicated that the total number of caterpillars in the spray-zones was reduced by 53.5% and 84.0% on snowberry and Garry oak, respectively. These estimates are considerably higher than those of 1999, which were 66.3% and 29.1% on snowberry and Garry oak, respectively. Thus, monitoring non-target Lepidoptera in 2000 was important and supported the hypothesis that the full extent of the pesticide side effects would be manifest only in the long-term.

The winter moth population appeared to recover partially between the years 2000 and 2003 but it still remained more than five times lower in the treatment plots than the check plots. This may be considered a beneficial side effect of the *Btk* applications since gypsy moth is a destructive and

non-native defoliator of Garry oak and numerous other trees and shrubs. As a group, the other non-target Lepidoptera on Garry oak remained reduced in the treatment plots by 52.9%, indicating that their recovery was incomplete at four years after the 1999 *Btk* applications. Nevertheless, there were several lines of evidence to indicate that by 2003 there was substantial recovery. For example, each of the 11 species that was initially harmed by the 1999 *Btk* applications showed an increase in the treatment plots relative to the check plots between 2000 and 2003. Seven of these species had also increased their abundance relative to 1999 (based on post-spray data). In 2003, the check plot samples contained only five more species than the treatment plot samples, whereas in 1999 and 2000 the difference was 16 species in each year, respectively. There were numerous species (16 in 1999, 17 in 2000, and 7 in 2003) that were collected in the check plots and absent from the treatment plot samples. The converse situation (i.e. species occurring in the treatment plot samples that were absent in the check plot samples) did not occur in 1999 but it did occur in 2000 for one species (*A. dorsalana*) and in 2003 for two species (*A. dorsalana* and *N. californiaria*). More evidence of recovery comes from the reduction in the number of significantly affected species in 2003 (3 species) relative to the previous years of this monitoring project (9 species in each of 1999 and 2000). However, it should be noted that the lower sampling effort in 2003 relative to the previous years, reduced the power of our statistical tests, which in turn would reduce the probability of detecting lingering treatment effects. Perhaps the best indication that the non-target Lepidoptera in the treatment plots underwent considerable rebuilding of their populations between the years 2000 and 2003 comes from the mean number of larvae collected per Garry oak branch, which increased during that period by less than half of one percent in the check plots, compared with a rise in the treatment plots of more than 400 percent.

Over the duration of this study there were 13 species that were collected during only one or two of the three years that we sampled. Each of these species was uncommon in our samples (i.e. fewer than 60 specimens per year). This proves an important point – the absence of a particular species in the treatment plot samples for one or two years does not indicate its removal from the sprayed area. Rather, it reflects the fact that many lepidopteran species are distributed sparsely over large landscapes, thus making it virtually impossible to find all of them, especially when they have been reduced by pesticide use.

Research Paper on the 2003 Field Season

Introduction

The European gypsy moth (*Lymantria dispar* (L.)) has become a widespread and destructive forest and agriculture pest in eastern North America since its introduction in the 1860's. In British Columbia, the gypsy moth has become a potential threat to deciduous trees and shrubs in forest, agricultural and urban settings [1]. Its population is therefore closely monitored. By the late 1990's, pheromone trapping on southern Vancouver Island indicated that the gypsy moth population was growing to an unacceptable level. Consequently, the British Columbia Ministry of Forests conducted an eradication campaign during the spring of 1999, which consisted of three aerial applications of *Bacillus thuringiensis* subsp. *kurstaki* (*Btk*).

Btk is a common bacterium used in various formulations, to suppress the spread of lepidopteran species such as the gypsy moth [2]. Since its introduction to the market place in 1970, the popularity of *Btk* has steadily increased as a result of its specificity to lepidoptera, minimal impacts on humans and wildlife, and short residual activity [3,4]. Although *Btk* is an attractive alternative to broad-spectrum synthetic insecticides there has been concern regarding its potential to harm non-target organisms [5]. Many lepidopterans are known to be physiologically susceptible to *Btk* formulations [6-8] and several field studies have demonstrated that aerial spraying of *Btk* is generally harmful to non-target Lepidoptera [9-13]. Thus, it was hypothesized that the 1999 gypsy moth eradication program on southern Vancouver Island would be harmful, at least in the short term, to susceptible native lepidopteran species.

In this context, the present monitoring program was initiated to measure the immediate impacts (1999-2000) of the eradication program on non-target Lepidoptera and to estimate the level of recovery that had occurred by the spring of 2003, four years after the *Btk* applications. The results of the first two years of the study (which were submitted as reports to the British Columbia Forest Practices Branch) showed that the species richness, diversity, and total abundance of Lepidoptera on Garry oak and snowberry branches was significantly lower ($p=0.05$) in the areas that were sprayed with *Btk* than surrounding unsprayed areas. A significant reduction of the less abundant

species was also demonstrated by pooling the infrequently collected species together and analyzing them as a group. Sixteen species were collected in sufficient numbers for statistical analysis, three of which did not appear to be harmed by the *Btk* treatments, two of which were already less common in the treatment plots than the check plots before the *Btk* spray applications, and eleven of which were significantly reduced in the treatment plots following the applications of *Btk*.

Several other field studies assessing the direct impact of *Btk* sprays on non-target Lepidoptera have concluded that *Btk* treatments, applied during early spring, can cause immediate reductions in the population density of some immature Lepidoptera. However, the statistical demonstration of a change following a pesticide treatment does not provide information about the ecological importance of the disturbance, which is, ultimately, the major environmental concern regarding the use of *Btk*. For example, the observed reductions of non-target Lepidoptera in this monitoring project had no apparent effect on songbird populations in Garry oak habitats within the spray zones [14]. Moreover, with short generation times, high fecundity and vagility, many lepidopteran species may recover relatively quickly through reproduction and/or immigration from adjacent unsprayed habitats [15-17]. Therefore, assessing recovery is a vital component of pesticide impact assessment and is the focus of this report.

Methods

***Btk* applications**

Btk (Foray 48B®) was applied to two spray-zones in the Greater Victoria area by fixed-winged aircraft, at a rate of 50 BIU/ha in 4.0L/ha. Both zones were sprayed on May 8, May 19, and June 8, 1999 between 05:30 and 07:00 hrs. Three additional zones located at Mill Bay, Nanaimo and Tssawassen were also sprayed with *Btk* during the gypsy moth eradication program but they were not examined in this study. Spray droplets on Kromekote® cards (Smart papers LLC, Hamilton, OH, USA) (two to four cards per site) confirmed spray penetration to the ground at all our treatment sites. No deposits were detected at check plots located outside the defined spray-zone boundaries.

Study sites and sampling procedures

Immature Lepidoptera were monitored on Garry oak on southern Vancouver Island during the spring season from 1999-2003. Garry oak is the only oak native to British Columbia where it occurs in two general habitat types: 'open meadow' with large healthy oaks, and 'rocky knoll' with typically gnarled and stunted oaks [18]. Garry oak study sites were selected in semi-natural parks and private estates according to the criteria reported by Sopuck et al. [14]. In 2003, a total of 20 Garry oak sites were sampled: 9 were located within the larger (12,203-ha) spray zone in Greater Victoria, and one site was within the smaller (602-ha) separate spray-zone in Brentwood Bay. The 10 study plots within the spray-zones were paired with 10 similar unsprayed check plots located outside the aerial spray boundaries.

Insect sampling was timed to coincide with the peak abundance of most species. There was a total of five sample dates (fewer than the previous years), each requiring 3 days to complete. The five sample dates began on 10 May, 15 May, 20 May, 28 May and 1 June. At each site, two randomly selected trees were sampled at each sampling date by vigorously shaking 1 branch per tree for 10 seconds with a 2m wooden pole. The dislodged caterpillars fell onto a 2 m x 3 m white nylon sheet that was outstretched on the ground directly beneath the branch. The larvae were removed from the sheet, placed into plastic zip-lock bags with oak foliage, and refrigerated to slow

their activity. All species were identified in the larval stage and photographed by Robert Duncan (Canadian Forest Service) within three days of collection. Garry oak branches were selected only if: 1) they were not sampled previously, 2) they were within 2 m of the ground, 3) their base diameter was 4-10 cm, and 4) they possessed 30-60 leaf clusters. Leaf clusters were counted on all branches to ensure that equal amounts of foliage were sampled in the treatment and control plots. The average number of leaf clusters per branch sampled was 47.56 in the treatment plots and 47.96 in the check plots.

Data Analysis

Lepidoptera abundance was quantified as the number of larvae per branch as opposed to number of larvae per unit of foliage because the number of leaf clusters sampled was not significantly different between the treatment and check plots. Species encountered 60 or fewer times throughout the 2003 season were recorded as 'uncommon species', and were pooled together and analyzed as a group. Two factor ANOVA models were used to examine the variation among treatments (sprayed and unsprayed) and sample periods (1 to 5) in species richness, total lepidoptera abundance (all species combined), and the abundance of 12 individual species (*Archips rosana*, *Chionodes trichostola*, *Choristoneura rosaceana*, *Epinotia emarginana*, *Operophtera brumata*, *Hydriomena nubilofasciata*, *Telphusa sedulitella*, *Caloptilia sanguinella*, *Ypsolopha cervella*, *Pandemis cerasana*, *Erranis tiliaria vancouverensi*, and *Othosia pacifica*).

Results

Throughout the 2003 season of the monitoring program, 6,790 lepidopteran larvae were collected from 200 Garry oak branches; 1,938 in the treatment plots and 4,852 in the check plots (Table 1). A total of 33 species was collected in 2003, with 26 species in the treatment plots and 31 species in the check plots. Seven species were unique to the check plots and two species were found only in the treatment plots. Thus, species composition was more equitable between the treatment and check plots in 2003 than in the previous years of this monitoring program. Species names were placed on all the specimens except for two different Noctuidae species, which could not be identified.

ANOVA models showed that the difference in mean abundance per branch between the check and treatment plots was statistically significant for total abundance (Table 2). Among the six most abundant species in the 2003 samples, only one (*O. brumata*) was significantly more abundant in the check plots than the treatment plots; and only one species (*E. emarginana*) was significantly more abundant in the treatment plots than the check plots. Among the remaining 31 species collected in 2003, only two (*Y. cervella* and *E. vancouverensis*) were significantly more abundant in the check plots than the treatment plots, and none were significantly more abundant in the check plots. Temporal effects significantly influenced the mean abundance of three species (*O. brumata*, *H. nubilofasciata*, and *C. rosaceana*), each of which reached the pupal stage stage before the end of the 2003 field season. The interaction between treatment and sample period was statistically significant for *O. brumata* and *H. nubilofasciata* but not for the other species or species groups (Table 2).

As in the previous years of this monitoring project, *O. brumata* was the numerically dominant species on Garry oak, accounting for 52.3% of total abundance from 10 May to 3 June, 2003. *O. brumata* reached its maximum abundance near the middle of May (Fig. 1) and by 28 May it had almost disappeared from the samples (due to pupation). *T. sedulitella*, which was the second most abundant species in 2003, accounted for 10.2% of the total Lepidoptera abundance. The date of peak abundance was variable among the different species and could not be estimated for most species due to variation in the samples (Figures 2-15). The abundance of the uncommon species

(i.e. those collected less than 60 times in 2003) generally increased during early May and from 20 May onward remained relatively stable (Fig. 16). The mean number of species per branch increased slightly over the duration of the 2003 field season (Fig. 17) as a result of a seasonal increase in species richness. The number of species collected at sample dates 1-5 was; 15, 21, 24, 24, and 27, respectively.

Table 1: Lepidopteran species composition on Garry oak in Greater Victoria, 10 May – 3 June, 2003, in treatment plots (Btk-sprayed) and check plots (unsprayed).

Family Species	Number of individuals			Percentage of total abundance
	Treatment plots	Check plots	Total	
Argyresthiidae				
<i>Argyrotaenia dorsallana</i>	1	0	1	0.01
Gelechiidae				
<i>Chionodes trichostola</i>	127	243	370	5.45
<i>Telphusa sedulitella</i>	311	383	694	10.22
Geometridae				
<i>Operophtera brumata</i>	618	2935	3553	52.3
<i>Hydriomena nubilofasciata</i>	91	135	226	3.33
<i>Lambdina fiscellaria sominiaria</i>	0	4	4	0.06
<i>Cyclophora dataria</i>	0	1	1	0.01
<i>Venusia pearsalli</i>	1	12	13	0.19
<i>Eupithecia</i> spp.	0	2	2	0.03
<i>Neocalcis californiaria</i>	1	0	1	0.01
<i>Erranis tiliaria vancouverensis</i>	4	61	65	0.96
<i>Hemithea aestivarea</i>	1	3	4	0.06
<i>Coniodes plumogeraria</i>	3	2	5	0.07
Gracillariidae				
<i>Caloptilia sanguinella</i>	144	171	315	4.64
Lasiocampidae				
<i>Phyllodesma americana</i>	0	2	2	.03
<i>Malacosoma c. pluviale</i>	0	3	3	.04
Noctuidae				
<i>Orthosia pacifica</i>	66	32	98	1.44
<i>Lithophane contenta</i>	13	16	29	0.43
<i>Catocala aholibah</i>	3	3	6	.09
<i>Egira cognata</i>	19	13	32	0.47
<i>Egira nr crucialis</i>	2	5	7	0.10
<i>Aseptis binotata</i>	6	5	11	0.16
Unknown	0	1	1	0.01
Unknown	0	1	1	0.01
Oecophoridae				
<i>Carcina quercana</i>	4	16	20	0.29
Olethreutidae				
<i>Spilonota ocellana</i>	21	12	33	0.49
<i>Epinotia emarginana</i>	228	92	320	4.71
Plutellidae				
<i>Ypsolopha cervella</i>	24	198	222	3.27
Tortricidae				
<i>Pandemis cerasana</i>	137	362	499	7.35
<i>Choristoneura rosaceana</i>	46	53	99	1.46
<i>Archips rosana</i>	38	53	91	1.34
<i>Decodes fragarianus</i>	11	11	22	0.32
<i>Ditula angustiorana</i>	18	23	41	0.60
Total	1,938	4,852	6,790	100

Table 2: Results of two factor ANOVA models for effects of treatment and sample period on total Lepidoptera abundance, individual species abundance, uncommon species abundance, and species richness (species per branch).

Species	Mean abundance per branch		F Value		
	Treatment	Check	<i>Btk</i> treatment	Sample date	<i>Btk</i> /sample date interaction
<i>Chionodes trichostola</i>	1.27	2.43	2.95	1.60	59.39
<i>Archips rosana</i>	0.38	0.53	1.13	0.95	1.73
<i>Telphusa sedulitella</i>	3.11	6.22	0.52	1.63	1.53
<i>Orthosia pacifica</i>	0.66	0.32	3.09	0.79	0.69
<i>Operophtera brumata</i>	6.18	29.35	17.04**	7.10**	3.20**
<i>Caloptilia sanguinella</i>	1.44	1.71	0.21	0.24	15.22
<i>Pandemis cerasana</i>	1.37	3.62	4.3*	0.59	0.47
<i>Epinotia emarginana</i>	2.28	0.92	8.5**	0.2	1.33
<i>Hydriomena nubilofasciata</i>	0.91	1.35	3.15	4.02*	3.68**
<i>Choristoneura rosaceana</i>	0.46	0.53	0.13	4.93**	3.57
<i>Ypsolopha cervella</i>	0.24	1.98	8.79**	1.14	1.13
<i>Erranis tiliaria vancouverensis</i>	0.04	0.61	7.96**	0.32	0.48
Uncommon taxa (pooled)	1.04	1.45	3.03	1.8	1.7
Total Lepidoptera abundance	19.38	48.63	17.48**	4.82**	1.23
Species richness	5.82	7.10	6.5*	2.39	2.41

1. Asterisks indicate $p \leq 0.05$ (*) and $p \leq 0.01$ (**).
2. Degrees of freedom for main effects and interaction in all models are as follows: treatment = 1, sample date = 4, and treatment x sample date = 4. Error degrees of freedom for each model = 99.

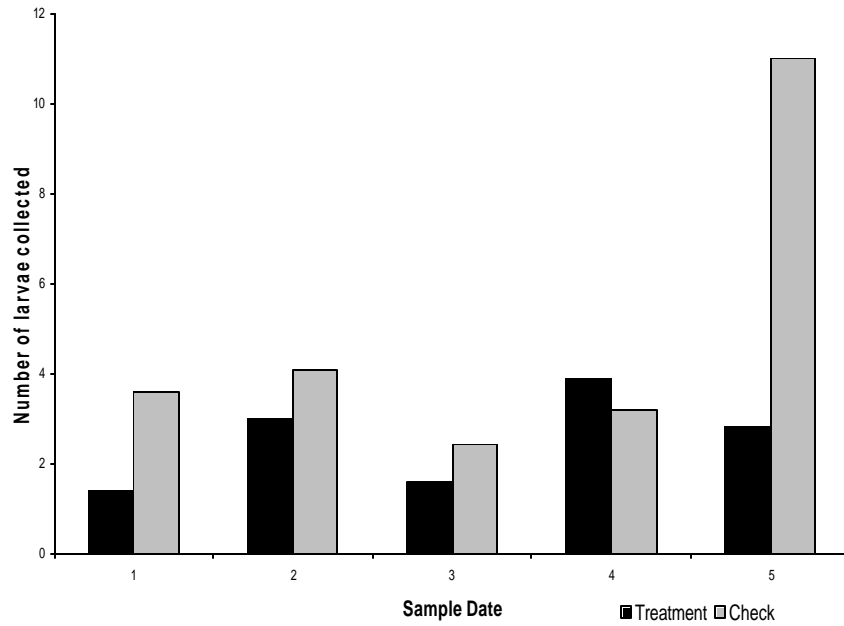


Figure 2: Abundance of *Chionodes trichostola* larvae (average number per 2 branches) collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

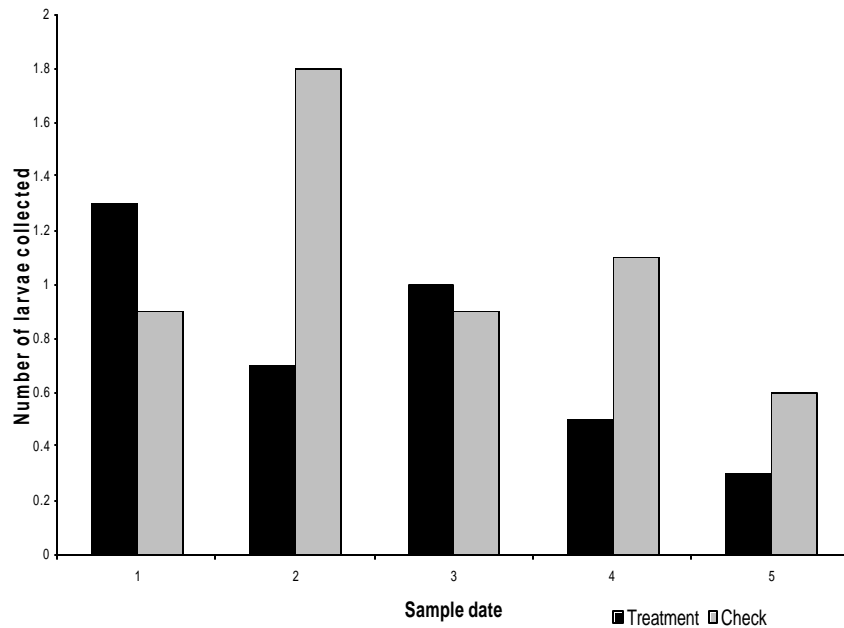


Figure 3: Abundance of *Archips rosana* larvae (average number per 2 branches) collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

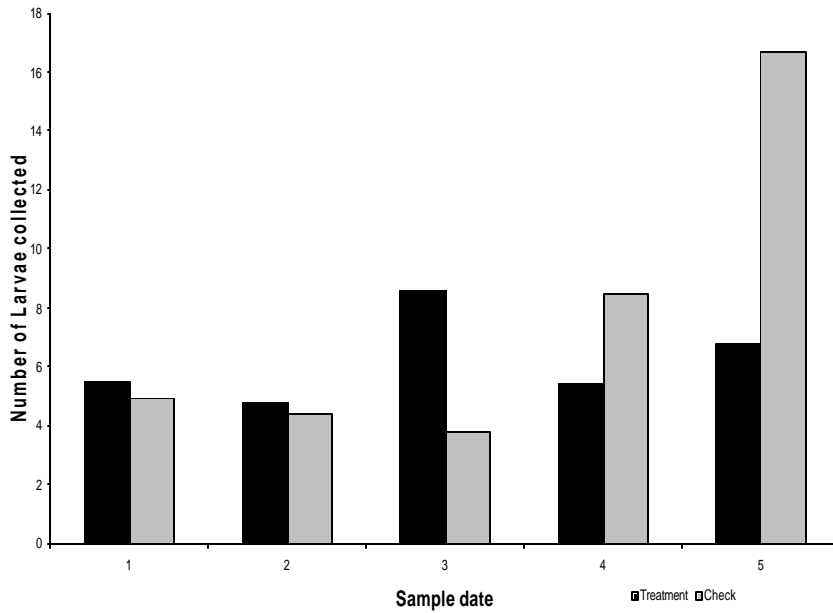


Figure 4: Abundance of *Telphusa sedulitella* larvae (average number per 2 branches) collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

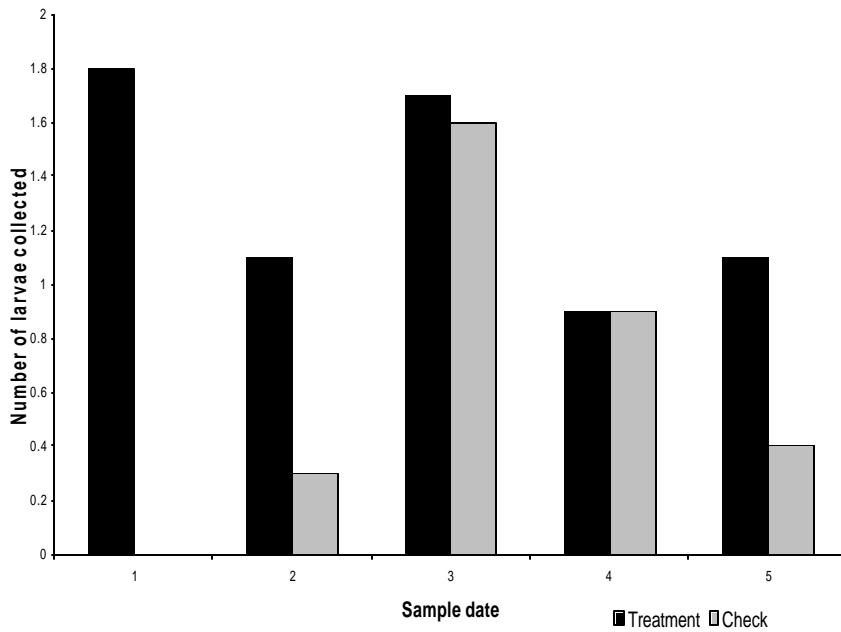


Figure 5: Abundance of *Orthosia pacifica* larvae (average number per 2 branches) collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

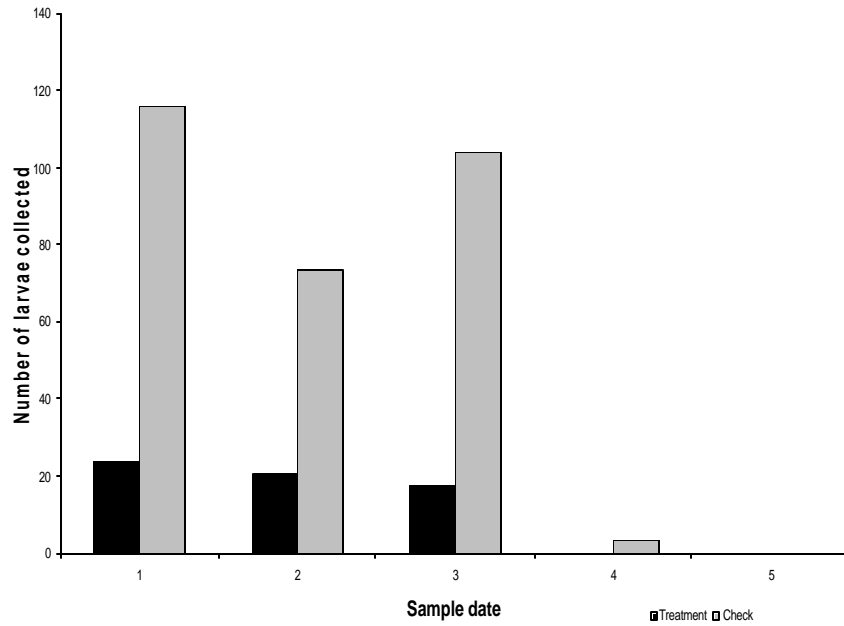


Figure 6: Abundance of *Operophtera brumata* larvae (average number per 2 branches) collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

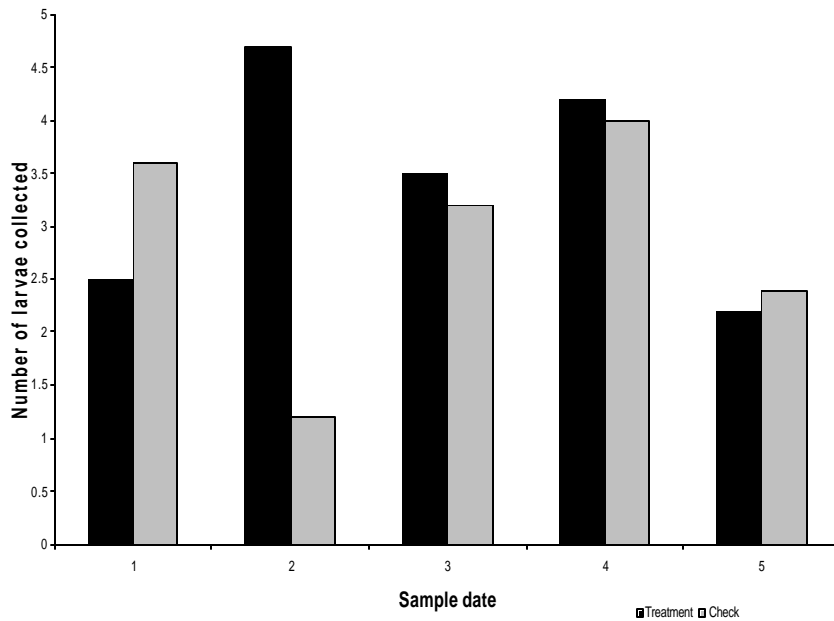


Figure 7: Abundance of *Caloptilia sanguinella* larvae (average number per 2 branches) collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

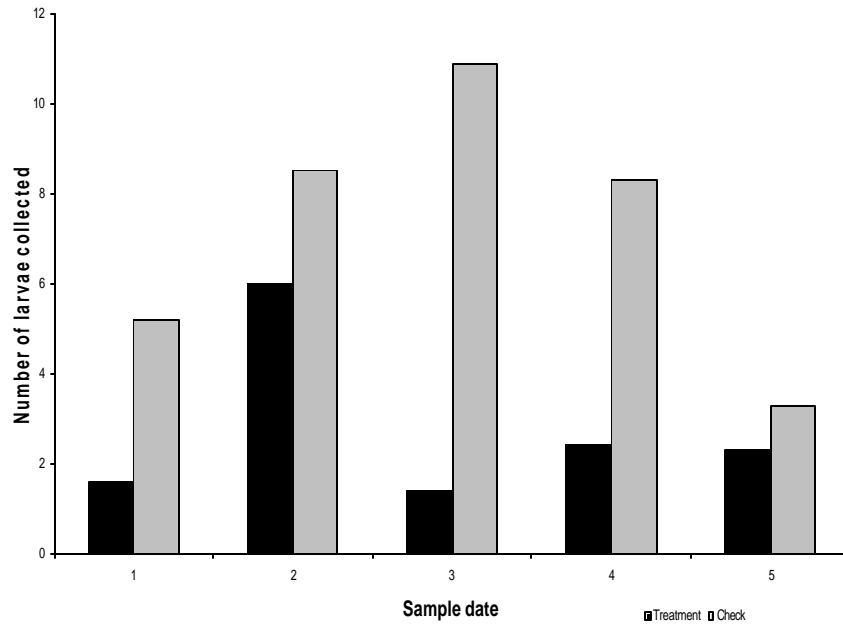


Figure 8: Abundance of *Pandemis cerasana* larvae (average number per 2 branches) collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

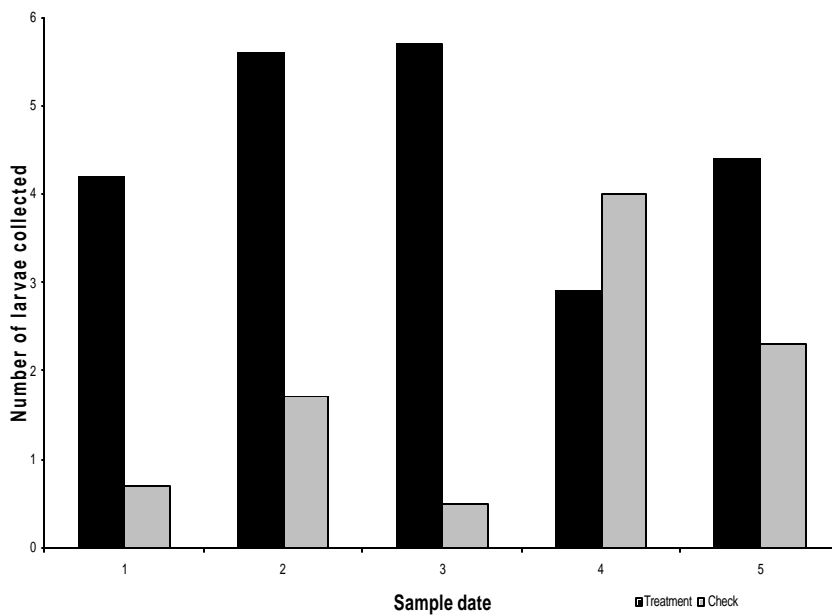


Figure 9: Abundance of *Epinotia emarginana* larvae (average number per 2 branches) collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

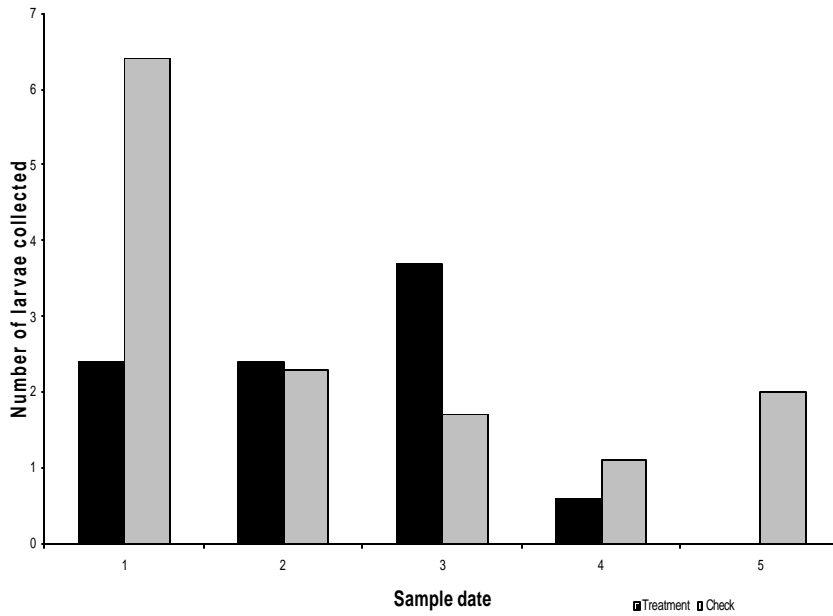


Figure 10: Abundance of *Hydiomena nubilofasciata* larvae (average number per 2 branches) collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

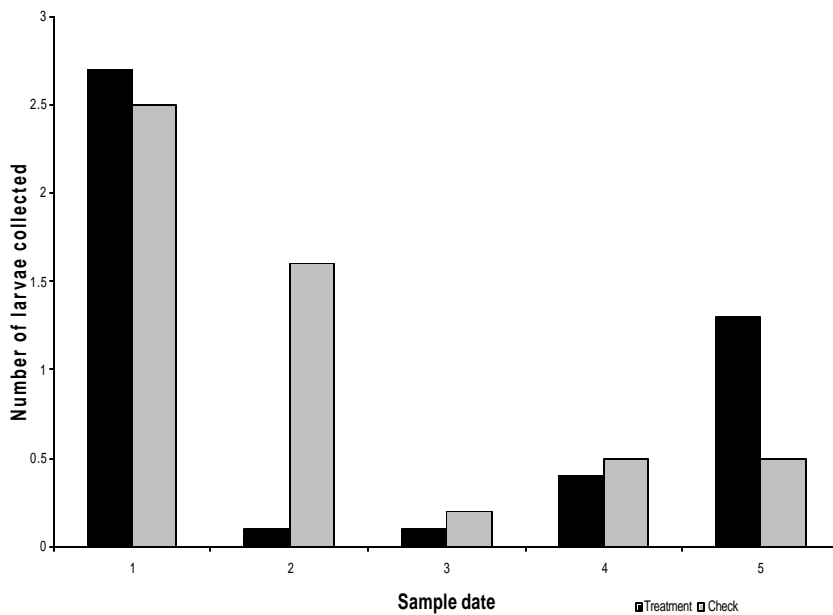


Figure 11: Abundance of *Choristoneura rosaceana* larvae (average number per 2 branches) collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

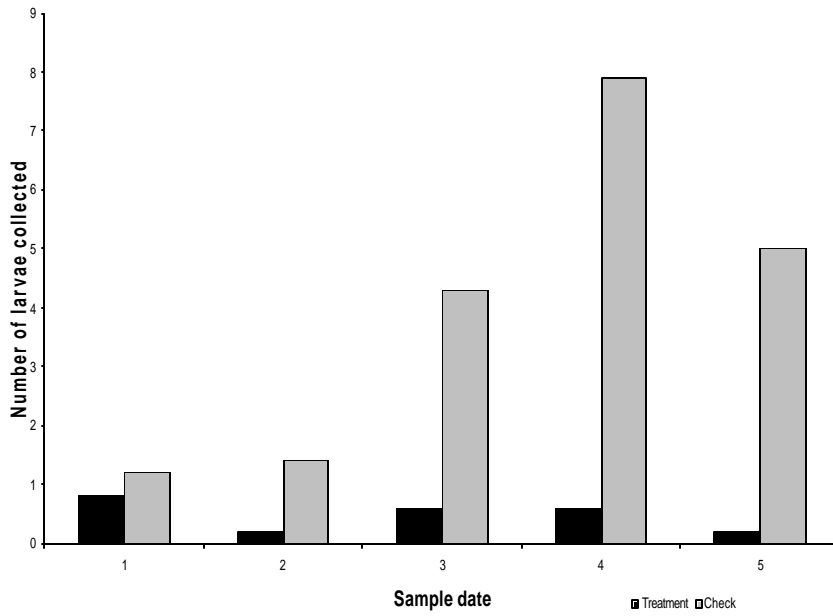


Figure 12: Abundance of *Ypsolopha cervella* larvae (average number per 2 branches) collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

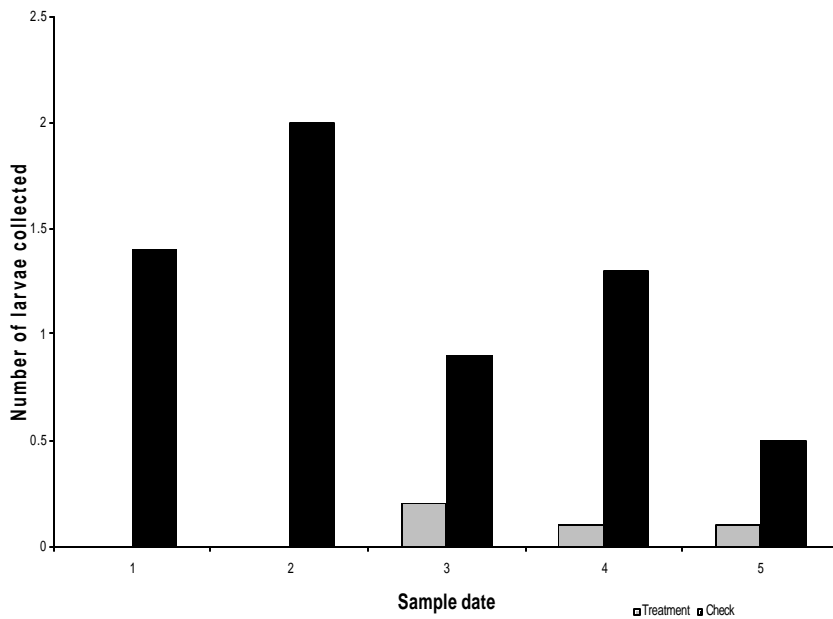


Figure 13: Abundance of *Erranis tiliaria vancouverensis* larvae (average number per 2 branches) collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

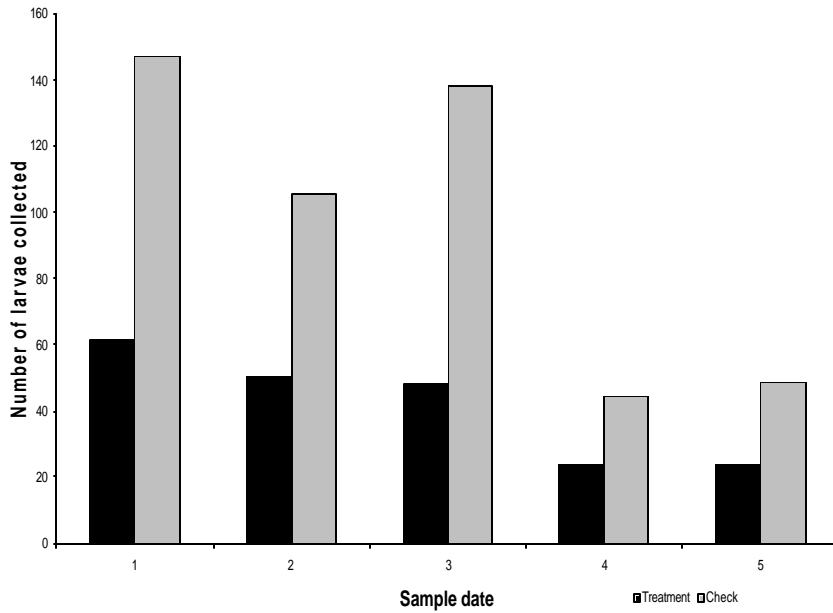


Figure 14: Total abundance (including winter moth) of immature Lepidoptera collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications (average number per 2 branches). The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

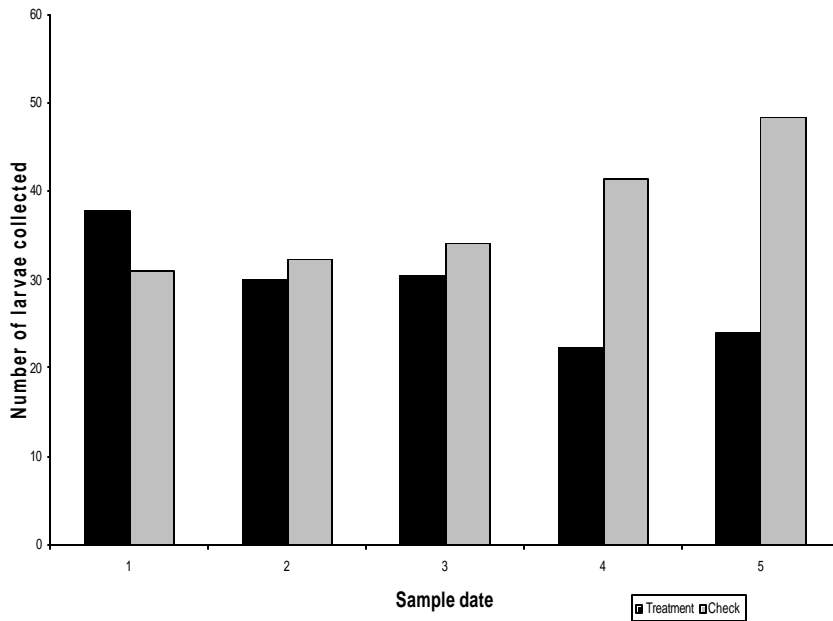


Figure 15: Total abundance (excluding winter moth) of immature Lepidoptera collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications (average number per 2 branches). The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

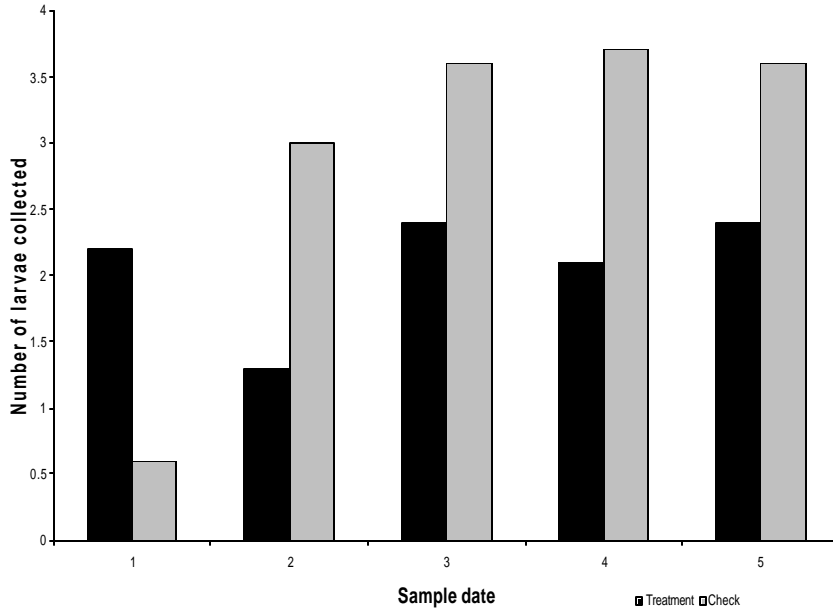


Figure 16: Abundance of the uncommon species (i.e those collected <50 times in total) from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

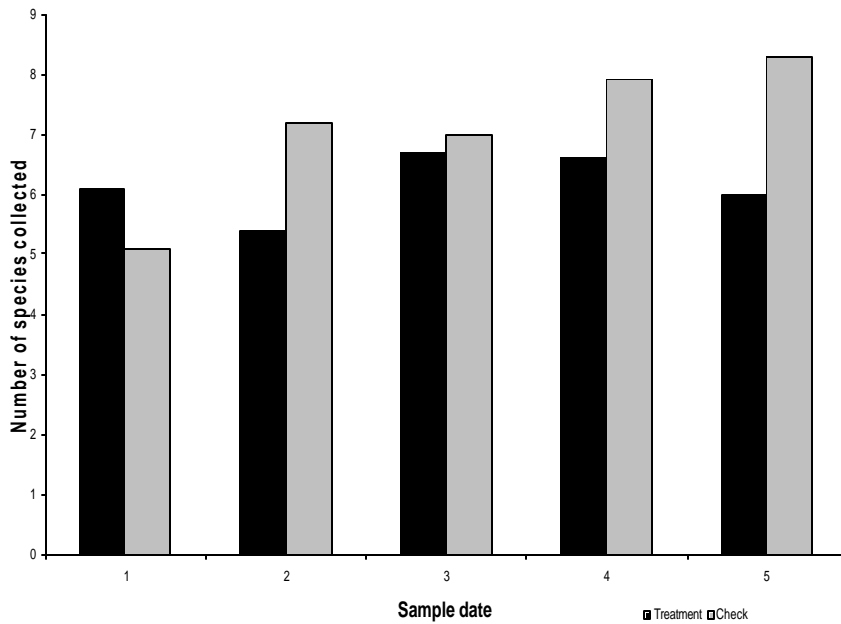


Figure 17: Mean species richness (average number of species per 2 branches) collected from Garry oak trees in the treatment and check plots during 2003, four years after the *Btk* applications. The sample dates were 10 May, 15 May, 20 May, 28 May and 1 June.

Discussion

Only three areas were sprayed with *Btk* on southern Vancouver Island in 1999, and only two of the sprayed areas contained Garry oak habitats that were suitable for this study. Therefore, strictly speaking, we had only two replicates in this study. However, the largest of the sprayed areas contained numerous Garry oak habitats, which we sampled and treated as replicates. Although our use of inferential statistics may constitute pseudoreplication [19] we believe that replication was approximated in this study because the treatment area was large and heterogeneous and each site in the treatment area was matched with a similar control site outside the spray area. In the future, data from this study may be combined with data from other studies involving non-target Lepidoptera on Garry oak [9,20] in a meta-analysis [21], resulting in a more powerful test of the null hypothesis.

For the vast majority of species there was a lack of significant difference between the treatment and check plots during the pre-spray period in 1999. This result should not be interpreted as evidence of equality among the plots because the pre-spray ANOVA models contained fewer samples and were, therefore, of lower power than the post-spray ANOVA models. Nevertheless, within 2 months of the *Btk* applications, the majority (9/14) of the abundant Lepidoptera species on Garry oak were significantly less abundant in the treatment plots than in the check plots. Moreover, the magnitude of the difference generally increased throughout the 1999 field season and reached a peak in 2000, one year after the *Btk* applications. In total, 11 species of non-target Lepidoptera on Garry oak were found to be reduced by the *Btk* applications (9 species in each of 1999 and 2000, and 5 species in 2003).

Based on the 1999 data, the total number of caterpillars on Garry oak in the spray-zones was reduced by an estimated 66.3%. The 2000 data indicated that the scale of the impact was considerably greater with an estimated reduction of 84.0%. These reductions may have been harmful to other animals that depend on immature Lepidoptera as a food source during the spring season. However, a concurrent study of birds [14] using 21 of our Garry oak sites, showed no impact of *Btk*-spraying on the relative abundance of most (41/42) adult birds. The spotted towhee (*Pipilo maculatus*) was the only potentially harmed bird, but in 2000 it was equally abundant in the

treated and check plots. This demonstrates that the percent mortality of Lepidoptera is less important to insectivorous birds than the absolute abundance of remaining individuals and the availability of alternative food. Whether or not other types of predators (i.e. small mammals) were harmed is unknown.

Four of the species (*O. brumata*, *H. nubilofasciata*, *C. rosaceana*, and *P. cerasana*) that were harmed by the *Btk* applications are non-native pests of Garry oak. Thus, reductions of these unwanted Lepidoptera may be considered a secondary benefit of the *Btk* spraying. Life history characteristics appeared to protect two of the unharmed species (*S. ocellana* and *C. trichostola*) in 1999. *S. ocellana* was not significantly reduced in the treatment plots in any year of the study, probably because it has an early phenology that caused it to reach the pupal stage before obtaining a lethal dose of *Btk*. *C. trichostola* remained common in the treatment plots throughout the 1999 field season, probably due to an extended period of larval emergence. In 2000 however, it was significantly less abundant in the treatment plots than the check plots indicating that it is susceptible to *Btk* residues in the field for up to 1 month. Likewise, Johnson et al. [6] demonstrated that *Btk* residues can remain toxic to sensitive species for at least 30 days under field conditions.

As a group, the uncommon (i.e. infrequently collected) species were significantly reduced by the *Btk* applications until at least 2001. In 2003, however, there was not a significant difference between the treatment and check plots in the abundance of the uncommon species group. Some of the uncommon species were probably still suppressed in the treatment plots in 2003 but with a paucity of information on the native moths, there is no way of knowing which ones would represent a risk to conservation (if any at all). In addition, it is probable that with a greater sampling effort in 2003 (i.e. larger sample sizes and more sample dates) a significant impact would have been detected for the uncommon species group. That said, there was partial recovery in the treatment plots by 2003, and for this reason (along with a relatively high sample variance) our samples did not detect significant differences for the uncommon species at the 95% confidence level.

More species were collected in the check plots than the treatment plots in each year of this monitoring program. Likewise, the average number of species per branch remained significantly higher in the check plots than the treatment plots until 2003. However, the apparent reduction of

lepidopteran species richness on Garry oak in the treatment sites following the *Btk* spray applications should be interpreted carefully because most species were collected infrequently. Species richness is a valuable parameter when all the non-target species of concern are collected frequently and can be identified easily, but the sampling effort necessary to achieve this may be impractical and possibly even harmful to local populations. Several authors [10-12] have reported that *Btk* causes immediate reductions in lepidopteran species richness, but their results may reflect diminished abundance rather than extirpation of sensitive species because species richness data are influenced by sample size, population density and species composition. Thus, species richness should be considered indicative rather than conclusive.

One way to compare the species richness data between the treatment plots and check plots, is to produce a treatment/check ratio, and then compare the ratios between years. For example, the average species richness per branch in 1999 (post-spray) was 7.03 species per branch in the treatment plots and 9.28 species per branch in the check plots, which can be expressed as the ratio ($7.03/9.28 = 0.76$). In the year 2000 there were considerably fewer species per branch in the treatment plots relative to the check plots, producing a ratio of 0.33. By 2003 however, the ratio increased to 0.87, indicating a general recovery of the affected species.

Several other lines of evidence point to the partial or complete recovery (by 2003) of most of the impacted species, but it must be stressed that the overall recovery was only partial, with an overall reduction in the treatment plots of 60.1% (52.9% excluding the winter moth). For example, each of the 11 species that was initially harmed by the 1999 *Btk* applications showed an increase in the treatment plots relative to the check plots between 2000 and 2003. Seven of these species had also increased their abundance relative to 1999 (based on post-spray data). In 2003, the check plot samples contained only five more species than the treatment plot samples, whereas in 1999 and 2000 the difference was 16 species in each year, respectively. There were numerous species (16 in 1999, 17 in 2000, and 7 in 2003) that were collected in the check plots and absent from the treatment plot samples. The converse situation (i.e. species occurring in the treatment plot samples that were absent in the check plot samples) did not occur in 1999 but it did occur in 2000 for one species (*A. dorsalana*) and in 2003 for two species (*A. dorsalana* and *N. californiaria*). More evidence of recovery comes from the reduction in the number of significantly affected species

in 2003 (3 species) relative to the previous years of this monitoring project (9 species in each of 1999 and 2000). However, it should be noted that the lower sampling effort in 2003 relative to the previous years, reduced the power of our statistical tests, which in turn would reduce the probability of detecting lingering treatment effects. Perhaps the best indication that the non-target Lepidoptera in the treatment plots underwent considerable rebuilding of their populations between the years 2000 and 2003 comes from the mean number of larvae collected per Garry oak branch, which increased during that period by less than half of one percent in the check plots, compared with a rise in the treatment plots of more than 400 percent. In the treatment plots, mean Lepidoptera abundance per branch was highest in 1999 (34.8), lowest in 2000 (3.35) and intermediate in 2003 (13.7). A slightly different trend occurred in the check plots, where Lepidoptera abundance was highest in 1999 (60.8), and almost the same in 2000 (18.6) and 2003 (18.7).

The total number of species collected on Garry oak branches in each year of the non-target Lepidoptera monitoring program was 52 in 1999 (this corrects an error in the 1999 report), 41 in 2000, and 33 in 2003. Over the duration of this study there were 13 species that were collected during only one or two of the three years that we sampled. Each of these species was uncommon in our samples (i.e. fewer than 60 specimens per year). This proves an important point – the absence of a particular species in the treatment plot samples for one or two years does not indicate its removal from the sprayed area. Rather, it reflects the fact that many lepidopteran species are distributed sparsely over large landscapes, thus making it virtually impossible to find all of them, especially when they have been reduced by pesticide use. Moreover, many species of Lepidoptera have populations that fluctuate widely from year to year, thus making them difficult to find in years of low abundance. That said, it is possible that the *Btk* treatments in 1999 extirpated one or more local populations of Lepidoptera but the present study does not provide any evidence to support such speculation. The use of light traps would make it easier to collect some of the sparsely distributed species and should be considered in future studies.

More than half of the known Lepidoptera fauna on Garry oak were not present in our samples, indicating that many of them are probably rare. Some of the species that we did encounter were probably also rare, particularly among the group of unidentified species. Rare species are of concern because they are thought to be more easily extirpated by disturbances like *Btk* spraying than common species [10]. We also collected several moth larvae that may be vulnerable in

Canada because their only food plant, Garry oak, is becoming increasingly threatened in British Columbia [22]. The reductions of non-target Lepidoptera observed in this study may impose further stress on Garry oak ecosystems on southern Vancouver Island. Natural Garry oak habitat has been almost completely destroyed in British Columbia due to urbanization, industrialization, agriculture and the invasions of exotic species [23]. The remaining fragments of the original habitat contain about 75% of British Columbia's endangered, threatened and vulnerable plant taxa, and they are considered to be at high risk with respect to biodiversity conservation [23]. One butterfly species (*Erynnis propertius*) and three subspecies (*Coenonympha californica insulana*, *Euchloe ausonides isulanus* and *Euphydryas editha taylori*) are found only in oak meadows and rocky knolls. Only *E. propertius* was present in our samples, and it was absent in 2003. *C. californica insulana* occurs on Vancouver Island but is red listed by the Province of British Columbia; *E. ausonides isulanus* is listed by COSEWIC as extirpated from Vancouver Island with only a single known population on San Juan Island; and *E. editha taylori* is listed by COSEWIC as endangered with no extant populations on Vancouver Island. Moths could also be at risk of becoming locally extinct, but there is a paucity of information on this large and diverse taxon.

Analysis of the non-target Lepidoptera monitoring program (1999-2003)

Initial impacts of *Btk* on non-target Lepidoptera

In general there was a lack of significant difference between Lepidoptera in the treatment and check plots during the pre-spray period in 1999, in terms of the abundance and species richness of larvae on snowberry and Garry oak. The only exceptions were *H. nubilofasciata* and *E. tiliaria vancouverensis*, which were more abundant on oak in the control sites during the pre-spray period, and *E. castella*, which was more abundant on snowberry in the treatment sites during the pre-spray period. Within 5 weeks of the 1999 *Btk* applications however, the uncommon species groups on snowberry and Garry oak were significantly more abundant in the control sites than the treatment sites, as were most of the common species. Lepidoptera species richness also dropped in sprayed plots relative to control plots on both host plant species. Therefore, I concluded that the 1999 gypsy moth eradication program on southern Vancouver Island was generally harmful to non-target Lepidoptera that were present in the feeding stage on snowberry and Garry oak when the *Btk* was applied. Moreover, since larvae were reduced on snowberry, a shrub with an early phenology, it appeared that the majority of Lepidoptera in the spray-zones were unable to complete their larval stage before the *Btk* spraying began.

The results in 2000, the second year of this monitoring program, showed that the impact of *Btk* on non-target Lepidoptera was more severe at 12-13 months post-spray than at 1-2 months post-spray. This lag time probably occurred because different species and different individuals of the same species varied in the amount of time they required to ingest a lethal dose of the pesticide. Some individuals may have ingested a lethal dose within a few hours of the first pesticide application and died within one to three days [24]. Other individuals may have ingested a lethal dose of *Btk* within days or even weeks of application [6]. As a result, the post-spray samples collected during 1999 contained individuals that would have died *in situ* (if they were not sampled) as a result of ingesting *Btk*. By 2000, however, the *Btk* was fully degraded (*Btk* formulations are not known to remain active to sensitive species in field environments beyond 30 days) [6]. Therefore, samples collected during 2000 reflect the scale of the *Btk* effect more accurately than

samples collected during 1999 because the 2000 samples did not contain individuals that were about to succumb to *Btk*.

At 12-13 months post-spray, four additional species (3 on Garry oak and 1 on snowberry) were shown to be reduced by the 1999 *Btk* applications, indicating that some effects went undetected in 1999. The data in 2000 also indicated that the total number of caterpillars in the spray-zones was reduced by 53.5% and 84.0% on snowberry and Garry oak, respectively. These estimates are considerably higher than those of 1999, which were 29% and 53% in the treatment and check plots, respectively. Thus, monitoring non-target Lepidoptera in 2000 was important and supported the hypothesis that the full extent of the pesticide side effects would be manifest only in the long-term.

The observed reductions of non-target Lepidoptera may have been harmful to other animals that depend on immature Lepidoptera as a food source during the spring. However, a concurrent study of birds [14] using 21 of our Garry oak sites, showed no impact of *Btk*-spraying on the relative abundance of most (41/42) adult birds. The spotted towhee (*Pipilo maculatus*) was the only potentially harmed bird, but in 2000 it was equally abundant in sprayed and unsprayed plots. This demonstrates that the degree of larval mortality is less important to predators than the absolute abundance of remaining individuals and the availability of alternative food items. Whether or not other types of predators (i.e. small mammals) were harmed is unknown.

Most of the lepidopteran species were collected infrequently throughout all years of the monitoring program, and more than half of the known Lepidoptera fauna on snowberry and Garry oak was absent in our samples. Thus it appears that many of the native lepidopteran species on these two host plants are uncommon (i.e. sparsely distributed) on southern Vancouver Island. The decline of the 'uncommon' species groups in the *Btk* spray-zones suggests that some of the rare species in this study were probably severely impacted by the *Btk* applications. Rare species are of special concern because they may be more easily extirpated by disturbances like *Btk* spraying than common species [10]. Nevertheless, conservation considerations should also include reductions of common species because they can contribute the most to certain ecosystem processes [25].

The apparent reduction of lepidopteran species richness on Garry oak in the treatment sites following the *Btk* spray applications should be interpreted carefully because, as stated previously, most species were collected infrequently. As a result, the reduction of species collected in the sprayed treatment areas probably reflects diminished abundance, rather than extirpation. Species richness is a valuable parameter when all the non-target species of concern are collected frequently and can be identified, but the sampling effort necessary to achieve this may be impractical and possibly even harmful to local populations. Several authors [10-12] have reported that *Btk* causes immediate reductions in lepidopteran species richness, but their results may reflect diminished abundance rather than extirpation of uncommon species. Thus, species richness data must be interpreted carefully because they are influenced by sample size and species composition [26]. In addition, species richness indices can be insensitive to local extinctions when they are analyzed statistically, and in some cases local extinctions may not be apparent in short-term studies [27]. For example, if a lepidopteran species is depleted to the point that adults have difficulty locating mates, then there could be a lag-time between the initial disturbance and the point of extirpation [28].

Recovery of non-target Lepidoptera

It was not possible to compare our data between years using inferential statistics because there were factors that could not be controlled (i.e. weather, cyclical fluctuations in populations, inadvertent variation in sampling technique) that may affect Lepidoptera abundance. Nevertheless, I have made several informative comparisons between the different years of the study, which are outlined in this section. The reader should keep in mind that only Garry oak was sampled in 2003 and there were fewer sample dates, sample sites, and sample branches per site than in the previous years of this study (Table 3). In addition, the dates of sampling varied between years (Table 4).

Table 3: Comparison of sample designs from 1999-2003 for monitoring non-target Lepidoptera on Garry oak.

Year	Number of sample dates	Number of sample sites	Number of sample branches
1999	7	28	588
2000	9	22	594
2003	5	20	200

Table 4: Comparison of sample dates from 1999 -2003 for monitoring non-target Lepidoptera on Garry oak.

Sample Period	1999	2000	2003
1	April 26-30	April 25-29	May 10-12
2	May 2-6	May 1-5	May 15-17
3	May 10-13	May 8-12	May 20-22
4	May 21-24	May 15-19	May 28-30
5	June 1-4	May 22-26	June 1-3
6	June 16-19	May 29 - June 2	
7	July 2-6	June 5-10	
8		June 12-16	
9		June 19-23	

In the treatment plots, mean Lepidoptera abundance per branch was highest in 1999 (34.8), lowest in 2000 (3.35) and intermediate in 2003 (13.7). A slightly different trend occurred in the check plots, where Lepidoptera abundance was highest in 1999 (60.8), and almost the same in 2000 (18.6) and 2003 (18.7). It is clear from Table 5 that in general, the affected non-target Lepidoptera in the treatment plots underwent considerable rebuilding of their populations between the years 2000 and 2003. During that period the mean number of larvae collected per Garry oak branch increased by less than half of one percent in the check plots, compared with a rise in the treatment plots of more than 400 percent.

Table 5: Mean number of lepidopteran larvae (excluding winter moth) collected in Greater Victoria in each year of the monitoring program. Mention ANOVA results in text.

<i>Sample Period</i>	<i>Mean number of larvae collected (per branch) in 1999</i>		<i>Mean number of larvae collected (per branch) in 2000</i>		<i>Mean number of larvae collected (per branch) in 2003</i>	
	Treatment	Check	Treatment	Check	Treatment	Check
1	24.4	27.9	3.18	9.93	15.0	15.5
2	38.0	52.9	0.40	11.85	14.9	16.2
3	39.9	73.2	0.97	10.78	15.3	17.0
4	75.1	88.4	2.76	18.51	11.2	20.7
5	32.3	100.2	4.48	23.36	12.0	24.1
6	18.7	36.8	3.82	24.01		
7	15.1	46.0	5.94	21.12		
8			3.61	22.12		
9			5.00	25.91		
Average	34.78	60.77	3.35	18.62	13.68	18.70

Measures of species richness are influenced by sampling size, sampling efficiency and species composition. Since these parameters were not held constant throughout the four years of this monitoring program, any comparison of species richness data between years must be interpreted carefully. One way to compare the species richness data between the treatment plots and check plots, is to produce a treatment/check ratio, and then compare the ratios between years. For example, the average species richness per branch in 1999 (post-spray) was 7.03 species per branch in the treatment plots and 9.28 species per branch in the check plots, which can be expressed as the ratio $(7.03/9.28 = 0.76)$. In the year 2000 there were considerably fewer species

per branch in the treatment plots relative to the check plots, producing a ratio of 0.33. By 2003 however, the ratio increased to 0.87, indicating a general recovery of the affected species (Table 6). Additional evidence of recovery comes from the fact that in 2003, the check plot samples contained only five more species than the treatment plot samples, whereas in 1999 and 2000 the difference was 16 species in each year, respectively.

Table 6: Mean number of lepidopteran species per Garry oak branch, collected in Greater Victoria in each year of the monitoring program. Mention ANOVA results in text. And state the treat/check ratio for each year since it is indicative.

Sample Period	Mean number of species collected (per branch) in 1999		Mean number of species collected (per branch) in 2000		Mean number of species collected (per branch) in 2003	
	Treatment	Check	Treatment	Check	Treatment	Check
1	7.2	7.0	0.6	4.3	6.1	5.1
2	7.6	8.3	1.2	4.4	5.4	7.2
3	9.0	10.3	1.2	6.8	6.7	7.0
4	9.9	11.0	4.0	8.3	6.6	7.9
5	7.8	11.8	2.2	9.2	6.0	8.3
6	4.3	7.1	3.2	7.8		
7	4.1	6.2	2.5	6.2		
8			2.3	5.9		
9			1.9	4.4		
Average	7.13	8.81	2.12	6.37	6.16	7.10

There were numerous species (16 in 1999, 17 in 2000, and 7 in 2003) that were collected in the check plots and absent from the treatment plot samples (Table 7). The converse situation (i.e. species occurring in the treatment plot samples that were absent in the check plot samples) did not occur in 1999 but it did occur in 2000 for one species (*A. dorsalana*) and in 2003 for two species (*A. dorsalana* and *N. californiaria*). This adds evidence that by 2003 there was substantial (albeit incomplete) recovery of the uncommon (i.e. sparsely distributed) species in the treatment plots. Further evidence of recovery is seen in Table 8, which shows a reduction in the number of significantly affected species in 2003 (3 species) relative to the previous years of this monitoring project (9 species in each of 1999 and 2000). However, it should be noted that the lower sampling effort in 2003 relative to the previous years, reduced the power of our statistical tests, which in turn would reduce the probability of detecting lingering treatment effects. Nevertheless, several other

lines of evidence point to the partial or complete recovery of most of the impacted species. For example, each of the 11 species that was initially harmed by the 1999 *Btk* applications showed an increase in the treatment plots relative to the check plots between 2000 and 2003 (Table 9). Seven of these species had also increased their abundance relative to 1999 (based on post-spray data).

Table 7: Lepidoptera collected from Garry oak branches that were unique to the check plots (i.e. not present in the treatment plots).

<i>Species unique to the check plots</i>		
<i>1999</i>	<i>2000</i>	<i>2003</i>
<i>C. albata</i>	<i>C. albata</i>	<i>L. somnaria</i>
<i>C. plumogeria</i>	<i>E. vancouverensis</i>	<i>C. dataria</i>
<i>H. aestivarea</i>	<i>P. plumogeria</i>	<i>Eupithecia spp.</i>
<i>N. californiaria</i>	<i>H. aestivarea</i>	<i>P. americana</i>
<i>E. propertius</i>	<i>E. propertius</i>	<i>M. pluviale</i>
<i>P. americana</i>	<i>P. americana</i>	<i>Unknowns (2 spp.)</i>
<i>O. badia</i>	<i>A. binotata</i>	
<i>A. binotata</i>	<i>C. quercana</i>	
<i>A. citrana</i>	<i>S. ocellana</i>	
<i>Unknowns (7 spp.)</i>	<i>Y. cervella</i>	
	<i>D. fragarianus</i>	
	<i>Unknowns (6 spp.)</i>	

Table 8: List of species that were significantly ($p=0.05$) less common in the treatment plots than the check plots on Garry oak foliage.

<i>Species</i>	<i>1999</i>	<i>2000</i>	<i>2003</i>
<i>O. brumata</i>	+	+	+
<i>Y. cervella</i>	+	+	+
<i>H. nubilofasciata</i>	+	+	
<i>T. sedulitella</i>	+	+	
<i>C. sanguinella</i>	+	+	
<i>P. cerasana</i>	+	+	
<i>D. fragarianus</i>	+		
<i>O. pacifica</i>	+		
<i>E. vancouverensis</i>	+		+
<i>E. emarginana</i>		+	
<i>C. rosaceana</i>		+	
<i>C. trichostola</i>		+	

Table 9: The proportion of immature Lepidoptera collected in the treatment plots vs. check plots. All species listed were significantly ($p=0.05$) harmed by the 1999 *Btk* spray applications.

Species	1999		2000		2003	
	Treatment	Check	Treatment	Check	Treatment	Check
<i>O. brumata</i>	30.0	70.0	1.3	98.7	17.4	82.6
<i>Y. cervella</i>	32.1	67.9	0.0	100	10.8	89.2
<i>H. nubilofasciata</i>	23.6	76.4	6.6	93.4	40.3	59.7
<i>T. sedulitella</i>	36.9	60.1	11.5	88.5	33.3	66.7
<i>C. sanguinella</i>	46.3	53.7	30.3	69.7	45.7	54.3
<i>P. cerasana</i>	24.5	75.5	5.9	94.1	27.5	52.5
<i>D. fragarianus</i>	13.3	86.7	0.0	100	50.0	50.0
<i>O. pacifica</i>	28.0	72.0	12.8	87.2	67.3	32.7
<i>E. vancouverensis</i>	24.1	75.9	0.0	100	6.2	93.8
<i>E. emarginana</i>	68.2	31.8	24.8	75.2	71.3	28.7
<i>C. rosaceana</i>	46.6	53.4	18.4	81.6	46.5	53.5

The total number of species collected on Garry oak branches in each year of the non-target Lepidoptera monitoring program was 52 in 1999 (this corrects an error in the 1999 report), 41 in 2000, and 33 in 2003 (Table 10). This decline is probably due to the reduction of sample sites in 2000 and 2003, and the shorter field season in 2003. Over the duration of this study there were 13 species that were collected during only one or two of the three years that we sampled (Table 11). Each of these species was uncommon in our samples (i.e. fewer than 60 specimens per year). This proves an important point – the absence of a particular species in the treatment plot samples for one or two years does not indicate its removal from the sprayed area. Rather it reflects the fact that many lepidopteran species are distributed sparsely over large landscapes, thus making it virtually impossible to find all of them, especially when they have been reduced by pesticide use. Moreover, many species of Lepidoptera have populations that fluctuate widely from year to year, thus making them difficult to find in years of low abundance. That said, it is possible that the *Btk* treatments in 1999 extirpated one or more local lepidopteran populations but the present study does not provide any evidence to support such speculation. The use of light traps would make it

easier to collect some of the sparsely distributed species and should be considered in future studies.

Table 10: Number of species (including unidentified morphotypes) collected on Garry oak in Greater Victoria in each year of the monitoring program. (Mention t-test in text of results)

Sample Period	Number of species collected in 1999		Number of species collected in 2000		Number of species collected in 2003	
	Treatment	Check	Treatment	Check	Treatment	Check
1	22	20	4	14	15	15
2	21	21	6	15	18	22
3	24	23	10	20	23	19
4	21	28	13	24	18	18
5	22	22	8	25	16	21
6	15	24	15	22		
7	22	26	10	24		
8			10	24		
9			9	17		
Total	36	52	24	40	26	31

Table 11: Species that were collected from Garry oak branches during only one or two of the three years that we sampled.

Species	Number of specimens collected		
	1999	2000	2003
<i>Biston cognataria</i>	5	0	0
<i>Coniodes plumogeraria</i>	11	0	5
<i>Pero sp.</i>	2	0	0
<i>Egira simplex</i>	49	0	0
<i>Biston cognataria</i>	5	0	0
<i>Orgia antiqua badia</i>	4	0	0
<i>Argyrotaenia dorsalana</i>	0	1	1
<i>Phigalia plumogeria</i>	0	1	0
<i>Lithophane contenta</i>	0	27	29
<i>Egira cognata</i>	0	16	32
<i>Egira nr crucialis</i>	0	10	7
<i>Betodes angustiorana</i>	0	12	0
<i>Malacosoma c. pluviale</i>	0	0	3

Beneficial side-effects of *Btk* (i.e reductions of pest species)

It was hypothesized that the 1999 gypsy moth eradication program would have a positive side-effect in terms of reducing other lepidopteran pests besides the gypsy moth. Indeed, evidence was found that the *Btk* treatments significantly reduced four non-native oak defoliators (*O. brumata*, *H. nubilofasciata*, *C. rosaceana*, and *P. cerasana*). The most destructive of these introduced defoliators in the Greater Victoria area is *O. brumata* (the winter moth), which is an introduced species that attacks numerous other trees and shrubs. Winter moth populations were not significantly different between the treatment and check plots prior to the *Btk* applications in 1999 (based on two sample dates). Following the treatments, however, the winter moth remained significantly and substantially reduced in the treatment plots relative to the check plots (Table 12). The estimated reduction in 2000, one year after the *Btk* applications, was 98.7% on Garry oak and 56.7% on snowberry. The winter moth population appeared to recover partially between the years 2000 and 2003 but it still remained more than four times lower in the treatment plots than the check plots. The cause of the dramatic decline of the winter moth in our year 2000 samples is uncertain. The winter moth population may have crashed in that year but it is also possible that the lower abundance was caused by a change of personnel in the year 2000 (presumably some workers will dislodge more larvae during beating than others). However within each year, the same person, who maintained a constant sampling efficiency throughout, sampled all the plots. Thus, the within year analyses are valid.

Table 12: Mean number of *O. brumata* (winter moth) larvae collected between 1 May and 24 May on Garry oak branches in Greater Victoria in each year of the monitoring program. Mention ANOVA results in text.

<i>Sample period</i>	<i>Mean number of O. brumata larvae per branch in 1999</i>		<i>Mean number of O. brumata larvae per branch in 2000</i>		<i>Mean number of O. brumata larvae per branch in 2003</i>	
	Treatment	Check	Treatment	Check	Treatment	Check
Early May	37.7	50.1	0.24	6.73	11.8	58.1
Mid-May	37.8	86.3	0.00	5.97	10.4	36.7
Late May	35.9	74.6	0.15	2.94	8.8	52.0
Average	37.13	70.33	0.13	5.21	10.33	48.93

Recommendations

Further monitoring is recommended until there has been a complete recovery of the non-target Lepidoptera in the spray zones. Ideally sampling would occur each spring, but sampling at intervals of two or three years would likely produce informative results. Moreover, ongoing monitoring of the Lepidoptera associated with Garry oak is needed in Greater Victoria to provide baseline data on the population dynamics of the native and non-native species. Had such information been collected prior to the present monitoring program, I would have been in a much better position to make firm conclusions. Thus, continued monitoring is vital for providing important knowledge for future research.

References

1. Dreistadt, S.H. and D.L. Dahlsten. 1989. Gypsy moth eradication in Pacific coast states: history and evaluation. *Bulletin of the Entomological Society of America* 35:13-19.
2. Otvos, I.S. and S. Vanderveen. 1993. Environmental report and current status of *Bacillus thuringiensis* var. *kurstaki* use for control of forest and agricultural insect pests. Forestry Canada and Province of British Columbia, Ministry of Forests, Victoria, B.C.
3. Betz, F.S., S.F. Forsyth and W.E. Stewart. 1990. Registration requirements and safety considerations for microbial pest control agents in North America. pp 4-10 In M. Laird, L.A. Lacey, and E.W. Davidson. *Safety of Microbial Insecticides*. CRC Press, Boca Raton, Florida.
4. Waage, J. 1995. The use of exotic organisms as biopesticides: some issues. In Hokkanen, H.M., and J.M. Lynch [eds.]. *Biological Control: Benefits and Risks*. Cambridge University Press.
5. Oorton, D. 1987. The case against forest spraying with the bacterial insecticide *Bt*. *Alternatives* 15: 28-35.
6. Johnson, K.S., J.M. Scriber, J.K. Nitao and D.R. Smitley. 1995. Toxicity of *Bacillus thuringiensis* var. *kurstaki* to three nontarget Lepidoptera in field studies. *Environmental Entomology* 24: 288-297.
7. Herms, C., D.G. McCullough, L.S. Bauer, R.A. Haack, D.L. Miller, N.R. Dubois. 1997. Susceptibility of the endangered karner blue butterfly (Lepidoptera: Lycaenidae) to *Bacillus thuringiensis* var. *kurstaki* used for gypsy moth suppression in Michigan. *The Great Lakes Entomologist* 30: 125-141.
8. Peacock, J.W., D.F. Schweitzer, J.L. Carter, and N.R. Dubois. 1998. Laboratory assessment of the effects of *Bacillus thuringiensis* on Native Lepidoptera. *Environmental Entomology* 27 (2): 450-457.
9. Miller, J.C. 1990. Field assessment of the effects of a microbial pest control agent on nontarget Lepidoptera. *American Entomologist* 36: 135-139.
10. Miller, J.C. 1990. Effects of a microbial insecticide, *Bacillus thuringiensis* *kurstaki*, on nontarget Lepidoptera in a spruce budworm-infested forest. *Journal of Research on the Lepidoptera* 29: 267-276.
11. Sample, B.E., L. Butler, C. Zivkovich, R.C. Whitmore and R. Reardon. 1996. Effects of *Bacillus thuringiensis* var. *kurstaki* and defoliation by gypsy moth on native arthropods in West Virginia. *The Canadian Entomologist*. 128: 573-592.

12. Wagner, D.L., J.W. Peacock, J.L. Carter and S.E. Talley. 1996. Field assessment of *Bacillus thuringiensis* on nontarget Lepidoptera. *Environmental Entomology* 25: 1444-1454.
13. Boulton, T.J., I.S. Otvos and R.A. Ring. 2002. Monitoring nontarget Lepidoptera on *Ribes cereum* to investigate side effects of an operational application of *Bacillus thuringiensis* subsp. *kurstaki*. *Environmental Entomology* 31 (5) 903-913.
14. Sopuck, L., K. Ovaska, and B. Whittington. 2002. Responses of songbirds to aerial spraying of the microbial insecticide *Bacillus thuringiensis* var. *kurstaki* (Foray 48B?) on Vancouver Island, British Columbia, Canada. *Journal of Environmental Toxicology and Chemistry* 21: 1664-1672.
15. Jepson, P.C. 1989. The temporal and spatial dynamics of pesticide side-effects on non-target invertebrates. pp. 95-124 In P.C. Jepson. *Pesticides and non-target invertebrates*. Intercept, Dorset, England.
16. Thacker, J.R.M. and P.C. Jepson. 1993. Pesticide risk assessment and non-target invertebrates: integrating population depletion, population recovery, and experimental design. *Bulletin of Environmental Contamination and Toxicology*. 51: 523-531.
17. Whaley, W.H., J. Anhold and B. Schaalje. 1998. Canyon drift and dispersion of *Bacillus thuringiensis* and its effects on select nontarget lepidopterans in Utah. *Environmental Entomology*. 27: 539-548.
18. Sudworth, G.B. 1908. *Forest Trees of the Pacific Slope*. U.S. Department of Agriculture, Forest Service. Pp. 283-278.
19. Hurlbert, S.H. 1984. Pseudoreplication and the design of ecological field experiments. *Ecological Monographs* 54: 187-211.
20. Duthie, M. 1994. Effects of *Bacillus thuringiensis* var. *kurstaki* on nontarget Lepidoptera. Co-op Work Term Report, The University of Victoria. 35pp.
21. Gurevitch, J. and L.V. Hodges. 1999. Statistical issues in ecological meta-analysis. *Ecology* 80: 1142-1149.
22. Guppy, C.S., J.H. Shepherd and N.G. Kondla. 1994. Butterflies and skippers of conservation concern in British Columbia. *Can. Field Nat.* 108: 31-40.
23. Erickson, W. 1996. Classification and interpretation of Garry oak (*Quercus garryana*) plant communities and ecosystems in southwestern British Columbia. M.Sc. Thesis, University of Victoria.
24. Gill, S.S., E.A. Cowles and P.V. Pietrantonio. 1992. The mode of action of *Bacillus thuringiensis* endotoxins. *Annual Review of Entomology* 37: 615-636.

25. Hammond, P.C. and J.C. Miller. 1998. Comparison of the biodiversity of Lepidoptera within three forested ecosystems. *Conservation Biology and Biodiversity* 91: 323-328.
26. Spellerberg, I.F. 1991. *Monitoring Ecological Change*. Cambridge University Press, New York. pp 93-130.
27. Pimental, D. 1992. Ecological effects of pesticides on nontarget species in terrestrial ecosystems. pp 171-190 In R.G. Tardiff. *Methods to Assess Adverse Effects of Pesticides on Nontarget Organisms*. John Wiley and Sons, Toronto.
28. Grieg-Smith, P.W. 1992. The acceptability of pesticide effects on nontarget arthropods. *Aspects of Applied Biology* 31: 121-132.

Appendix A: Table of species composition for 1999.

Family <i>species</i>	Number of individuals			Proportion of Total Abundance (%)
	Treatment	Check	Total	
Arctiidae				
<i>Cleminsia albata</i>	0	1	1	0.01
Gelechiidae				
<i>Chionodes trichostola</i>	781	1354	2135	13.16
<i>Telphusa sedulitella</i>	412	706	1118	6.89
Geometridae				
* <i>Operophtera brumata</i>	1969	4571	6540	40.33
* <i>Hydriomena nubilofasciata</i>	384	1239	1623	10.01
<i>Erranis tiliaria vancouverensis</i>	42	132	174	1.07
<i>Venusia pearsalli</i>	12	34	46	0.28
<i>Coniodes plumogeraria</i>	0	11	11	0.07
<i>Cyclophora dataria</i>	6	1	7	0.04
** <i>Biston cognataria</i>	3	2	5	0.03
<i>Eupithecia</i> spp.	1	3	4	0.02
* <i>Lambdina fiscellaria somniaria</i>	1	3	4	0.02
** <i>Pero</i> sp.	1	1	2	0.01
*** <i>Hemithea aestivarea</i>	0	1	1	0.01
<i>Neocalcis californiaria</i>	0	2	2	0.01
unknowns (16 morphotypes)	27	38	65	0.40
Gracillariidae				
<i>Caloptilia sanguinella</i>	430	499	929	5.73
Hesperidae				
** <i>Erynnis propertius</i>	0	1	1	0.01
Lasiocampidae				
<i>Phyllodesma americana</i>	0	3	3	0.02
Lymantriidae				
<i>Orgia antiqua badia</i>	0	4	4	0.02
Noctuidae				
<i>Orthosia pacifica</i>	23	59	82	0.51
<i>Catocala aholibah</i>	25	35	60	0.37
<i>Egira simplex</i>	24	25	49	0.30
<i>Aseptis binotata</i>	0	3	3	0.02
unknowns (6 morphotypes)	7	7	14	0.09
Oecophoridae				
<i>Carcina quercana</i>	8	13	21	0.13
Olethreutidae				
<i>Spilonota ocellana</i>	818	873	1691	10.43
<i>Epinotia emarginana</i>	15	7	22	0.14
Plutellidae				
<i>Ypsolopha cervella</i>	109	228	337	2.08
Tortricidae				
* <i>Choristoneura rosaceana</i>	125	143	268	1.65
* <i>Archips rosana</i>	212	244	456	2.81
* <i>Pandemis cerasana</i>	74	228	302	1.86
* <i>Ditula angustiorana</i>	78	77	155	0.96
<i>Decodes fragarianus</i>	8	52	60	0.37
<i>Argyrotaenia citrana</i>	2	2	4	0.02
unknowns (1 morphotype)	0	1	1	0.01
Unidentified	10	8	18	0.11
Total	5607	10611	16218	100.00

* identifies introduced species and pests; ** identifies rare and/or potentially vulnerable species, *** indicates new record for Garry oak

Appendix B: Table of species composition for 2000.

Family Species	Number of Individuals			Proportion of Total Abundance (%)
	Treatment	Check	Total	
Arctiidae				
<i>Cleminsia albata</i>	0	1	1	0.01
Argyresthiidae				
<i>Argyrotaenia citrana</i>	1	3	4	
<i>Argyrotaenia dorsalanta</i>	1	0	1	0.01
Gelechiidae				
<i>Chionodes trichostola</i>	489	2731	3220	48.22
<i>Telphusa sedulitella</i>	102	788	890	13.33
Geometridae				
<i>Operophtera brumata</i>	11	842	853	12.77
<i>Hydriomena nubilofasciata</i>	6	85	91	1.36
<i>Lambdina fiscellaria sominiaria</i>	3	13	16	0.24
<i>Cyclophora dataria</i>	4	8	12	0.18
<i>Venusia pearsalli</i>	3	10	13	0.19
<i>Eupithecia</i> spp.	1	9	10	0.15
<i>Neoalcis californiaria</i>	2	1	3	
<i>Erranis tiliaria vancouverensis</i>	0	2	2	
<i>Phigalia plumogeria</i>	0	1	1	0.01
<i>Hemithea aestivareia</i>	0	1	1	0.01
Unknown	0	10	10	0.15
Gracillariidae				
<i>Caloptilia sanguinella</i>	153	352	505	7.56
Hesperiidae				
<i>Erynnis propertius</i>	0	1	1	0.01
Lasiocampidae				
<i>Phyllodesma americana</i>	0	1	1	0.01
Noctuidae				
<i>Orthosia pacifica</i>	5	34	39	0.58
<i>Lithophane contenta</i>	16	21	37	0.55
<i>Catocala aholibah</i>	3	19	22	0.33
<i>Egira cognata</i>	6	10	16	0.24
<i>Egira nr crucialis</i>	1	9	10	0.15
<i>Aseptis binotata</i>	0	1	1	0.01
Unknown	0	5	5	0.07
Oecophoridae				
<i>Carcina quercana</i>	0	32	32	0.48
Olethreutidae				
<i>Spilonota ocellana</i>	0	26	26	0.40
<i>Epinotia emarginana</i>	51	155	206	3.08
Plutellidae				
<i>Ypsolopha cervella</i>	0	61	61	0.91
Syrphidae				
Unknown	3	10	13	0.19
Tortricidae				
<i>Pandemis cerasana</i>	24	381	405	6.06
<i>Choristoneura rosaceana</i>	14	62	76	1.14
<i>Archips rosana</i>	13	22	35	0.52
<i>Decodes fragarianus</i>	0	15	15	0.22
<i>Ditula angustiorana</i>	3	11	14	0.21
<i>Betodes angustiorana</i>	1	11	12	0.18
Unknown	3	6	9	0.13
Unidentified	0	9	9	0.13
Total	919	5759	6678	99.79 ²

Appendix C: Table of species composition for 2003.

Family <i>Species</i>	Number of individuals			Percentage of total abundance
	Treatment plots	Check plots	Total	
Argyresthiidae				
<i>Argyrotaenia dorsallana</i>	1	0	1	0.01
Gelechiidae				
<i>Chionodes trichostola</i>	127	243	370	5.45
<i>Telphusa sedulitella</i>	311	383	694	10.22
Geometridae				
<i>Operophtera brumata</i>	618	2935	3553	52.3
<i>Hydriomena nubilofasciata</i>	91	135	226	3.33
<i>Lambdina fiscellaria sominiaria</i>	0	4	4	0.06
<i>Cyclophora dataria</i>	0	1	1	0.01
<i>Venusia pearsalli</i>	1	12	13	0.19
<i>Eupithecia</i> spp.	0	2	2	0.03
<i>Neoalcis californiaria</i>	1	0	1	0.01
<i>Erranis tiliaria vancouverensis</i>	4	61	65	0.96
<i>Hemithea aestivarea</i>	1	3	4	0.06
<i>Coniodes plumogeraria</i>	3	2	5	0.07
Gracillariidae				
<i>Caloptilia sanguinella</i>	144	171	315	4.64
Lasiocampidae				
<i>Phylodesma americana</i>	0	2	2	.03
<i>Malacosoma c. pluviale</i>	0	3	3	.04
Noctuidae				
<i>Orthosia pacifica</i>	66	32	98	1.44
<i>Lithophane contenta</i>	13	16	29	0.43
<i>Catocala aholibah</i>	3	3	6	.09
<i>Egira cognata</i>	19	13	32	0.47
<i>Egira nr crucialis</i>	2	5	7	0.10
<i>Aseptis binotata</i>	6	5	11	0.16
Unknown	0	1	1	0.01
Unknown	0	1	1	0.01
Oecophoridae				
<i>Carcina quercana</i>	4	16	20	0.29
Olethreutidae				
<i>Spilonota ocellana</i>	21	12	33	0.49
<i>Epinotia emarginana</i>	228	92	320	4.71
Plutellidae				
<i>Ypsolopha cervella</i>	24	198	222	3.27
Tortricidae				
<i>Pandemis cerasana</i>	137	362	499	7.35
<i>Choristoneura rosaceana</i>	46	53	99	1.46
<i>Archips rosana</i>	38	53	91	1.34
<i>Decodes fragarianus</i>	11	11	22	0.32
<i>Ditula angustiorana</i>	18	23	41	0.60
Total	1,938	4,852	6,790	100