Abstract: We analyzed the reproductive success of eastern bluebirds (Sialia sialis) nesting in apple orchards and non-orchard nest-boxes in southern Ontario, Canada, from 1988 to 2018. Using data from 2397 nest-boxes monitored at 20 orchard sites and 52 non-orchard sites, we first modeled phenological parameters typically linked to climate change across both site types. We found that the first egg of each brood was laid significantly earlier in the season each year over our 31-year study. Clutch initiation occurred 4 days earlier in the spring in 2018 compared to 1988. Average clutch size in the first or second brood did not change significantly during our 31-year study; however, clutches were significantly smaller in orchards compared to non-orchards (0.10 ± 0.03 fewer eggs between sites). Nests built in orchards were also at 6.1-fold greater risk of parasitism and 2.1-fold greater risk of depredation than nests in non-orchards. After accounting for depredation and nest parasitism, hatching success was still significantly lower in orchards than in non-orchards. Overall, hatching success was 4%–5% lower in orchards. The probability of successfully fledging did not differ significantly between site types. In 2012, a ban on use of the organophosphate insecticide azinphos-methyl in orchards was enacted in Canada. We did not find a difference in hatching or fledging success in orchards after the ban. In our assessment of available data, we conclude that any pesticide effect on hatching success of eastern bluebirds in sprayed orchards is most likely the consequence of long-term exposure to dichlorodiphenyltrichloroethane (DDT) compounds in orchard soils and bioaccumulation in eggs rather than pesticides in use since regulation of DDT in the 1970s. Environ Toxicol Chem 2021;40:3369–3378. © 2021 Her Majesty the Queen in Right of Canada. Environmental Toxicology and Chemistry published by Wiley Periodicals LLC on behalf of SETAC. Reproduced with the permission of the Minister of Environment and Climate Change Canada.

Keywords: Eastern bluebirds; Azinphos-methyl insecticides; Songbird reproductive success; Agricultural habitats

INTRODUCTION

Land use and climate change are the two leading threats to biodiversity in terrestrial ecosystems (Newbold et al., 2015; Sala et al., 2000; Urban, 2015). Globally, it is projected that by the year 2050, 48–52 land bird species will become extinct because of these anthropogenic factors (Jetz et al., 2007), and >80% of threatened birds are imperiled by conversion of habitat into agricultural land (Tilman et al., 2017). Effects of agriculture on grassland birds are particularly severe, due to habitat loss and range reduction (Jetz et al., 2007; Rosenberg et al., 2019). The quality of the remaining habitat available to wildlife, much of it agricultural, may be compromised by the effects of agrochemicals on non-target vertebrates and invertebrates alike (Eng et al., 2019; Goulson, 2013; Gress et al., 1973; Grier, 1982). In addition to the effects of agrochemicals, agricultural practices like tilling, mowing, and haying significantly reduce the reproductive success of nesting birds (Grüebl et al., 2015; Perlut et al., 2006).

Long-term landscape ecology studies are effective for testing relationships between climate change (Carleton et al., 2019; Chase et al., 2005; Dunn & Winkler, 1999) or habitat conversion (Crocker & Lawrence, 2018; Graham & DesGranges, 1993; Rattner et al., 2018) and avian reproductive success. Those studies are particularly valuable for grassland songbirds because their reproductive success is impaired by changes to migratory and breeding phenology (Cotton, 2003; Jones & Cresswell, 2010; Shipley et al., 2020). However, few studies span multiple
generations. We present a study of the reproductive success of eastern bluebirds (Sialia sialis) in conventionally sprayed apple orchard sites and non-sprayed, non-orchard sites spanning more than three decades (1988–2019).

Within our orchard sites, a wide variety of agrochemicals including fungicides, herbicides, and insecticides were applied annually throughout the study (Ontario Ministry of Agriculture, Food and Rural Affairs, 2021). In particular, the organophosphate insecticide azinphos-methyl was applied annually until 2012, when it was no longer available for use on apple orchards in Canada (Food and Agriculture Organization of the United Nations, 2013). This regulatory change was made partly because of the acute neurotoxicity of organophosphate insecticides to vertebrates (Bishop, Boermans, et al., 1998; Bishop, Van Der Kraak, et al., 1998; Bunyan et al., 1968, 1969; Elliott et al., 1996; Graham & DesGranges, 1993; Hill & Fleming, 1982). Sublethal and lethal neurotoxic effects have occurred in songbirds exposed to azinphos-methyl in agricultural habitats (Graham & DesGranges, 1993; Grue et al., 1997) including depression of cholinesterase in eastern bluebirds (Burgess et al., 1999) in the orchard study sites of the present study. Moreover, organochlorine pesticides including dichlorodiphenyltrichloroethane (DDT) were also used from the late 1940s until the early 1970s in apple orchards in Canada and were measured at concentrations >100 µg/g (wt wt) in eastern bluebird eggs in the 1990s (Bishop et al., 2000) and in American robin (Turdus migratorius) eggs as recently as 2019 (Kesic et al., 2021).

Bluebirds are open-woodland cavity-nest adopters and readily nest in human-made boxes, making them useful models for long-term landscape ecology studies because characteristics of nest location and clutch size can be experimentally controlled among individuals (Bauldry et al., 1995). For our study, bluebirds inhabited nest-boxes in orchard or non-orchard site types in southern Ontario, Canada. At this latitude (~43°N), bluebirds are migratory and multibrooded (Gowaty & Pliisner, 2020). In the northern parts of their range, mean clutch size decreases linearly by date and brood number (Dhondt et al., 2002).

We predicted that orchard nest-box sites would yield lower reproductive success for bluebirds. We therefore investigated characteristics of the habitat that influence reproduction. The first objective of our study was to test the effects of changing local climate during our multidecade study on the phenology of eastern bluebirds; our second aim was to identify biotic influence reproduction. The first objective of our study was to test the effects of changing local climate during our multidecade study on the phenology of eastern bluebirds; our second aim was to identify biotic factors that may affect nesting success differently in orchard and non-orchard sites, with a third objective to determine if there was a difference in reproductive success (hatching and fledging success) between orchard and non-orchard nest sites. Finally, we tested whether the ban on the use of the insecticide azinphos-methyl in 2012 significantly altered the probability of successful hatching or fledging in orchard sites.

**METHODS**

**Study sites and nest box monitoring**

We analyzed 2397 nest-boxes monitored and maintained by the principal author (W. F. Read) between April 17, 1988, and July 14, 2018, at 22 orchard sites and 61 non-orchard sites (Supporting Information, Table S1). Sites were located in the Great Lakes basin region of southern Ontario (approximate latitude 43°, longitude 80°; Supporting Information, Table S2). All monitored nest-boxes were located 8–10 m from the nearest neighbor. In orchard sites, nest-boxes were erected between orchard trees. Axle grease was applied to the lower half of all nest-box posts to deter mammalian predators. Nest-boxes were installed facing south, to avoid effects of the prevailing wind from the northwest and east. Openings to nest-boxes in orchard sites were oriented perpendicular to the lanes between tree lines, to minimize the probability of agrochemical sprays directly entering the boxes.

Boxes were checked every 3 days during the expected egg-laying period (expected dates for egg laying were determined from personal observation), every 2 days during hatching, and every 4 days during incubation and fledging periods. At each nest observation, the numbers of eggs, young (live or dead), and fledged chicks were recorded. Active nests were visited until the fates of all offspring were determined. The fate of an egg was considered “unhatched” when chicks of the same brood reached an age of 2 days. Ages of nestlings on first observation were estimated using established developmental characteristics (Gowaty & Pliisner, 2020) and comparison to other young of known age. Date of first egg (clutch initiation) was recorded either by observation of the first egg in the nest or by back-dating from hatching dates.

**Pesticide spray exposures**

The conventionally sprayed orchards in the present study were sprayed with a wide variety of pesticides during 1988–2018. The sprayed orchard and non-orchard sites were the same as those used in related studies where spray schedules from growers were collected in 1988–2001 and reported as types and spray application rates in Bishop et al. (2000) and Mayne et al. (2004; summarized in Supporting Information, Table S3). Also from 1988 to 1994, DDT and its metabolites were measured in eggs of tree swallows (Tachycineta bicolor), eastern bluebirds (S. sialis), and American robins (Turdus migratorius) nesting in conventionally sprayed orchards used in the present study (Bishop et al., 2000; Harris et al., 2000). In orchards, p,p’-dichlorodiphenyltrichloroethane (p,p’-DDE) was detected in the eggs at concentrations of 0.38–2.56 µg/g in tree swallow eggs and 0.37–105.1 µg/g in eastern bluebird eggs, whereas eggs from the same species sampled in one of our reference sites contained <0.6 µg/g p,p’-DDE (Bishop et al., 2000). Studies also confirmed the use of azinphos-methyl for codling moth (Cydia pomonella) control during that period in all the conventionally sprayed orchards (Bishop et al., 2000; Harris et al., 2000). In 2000–2001, mean p,p’-DDE concentrations ranged from 6.18 to 48.11 µg/g in eastern bluebird eggs sampled from four orchards (Mayne et al., 2004), which were also among those sampled in our study. During 2002–2018, the principal author (W. F. Read) observed that pesticide use continued at all of the conventionally sprayed orchards and in discussion with orchard growers confirmed
that their spray schedules conformed to the pesticide sprays recommended by the Ontario provincial government in those years (Ontario Ministry of Agriculture, Food and Rural Affairs, 2018, 2021). Similar observations for conventionally sprayed apple orchards were confirmed by other researchers in 2018 within this same area of Ontario, where >40 insecticides, miticides, and fungicides were used in apple orchards (Blechschmidt, 2020).

**Statistical analyses**

Of the 2397 nest-boxes observed between April 17, 1988, and July 14, 2018 (n = 21–113 nests/year across all sites; n = 2–73 non-orchards and n = 16–96 orchards; Supporting Information, Table S1), 2310 nests were included in our statistical analyses; five nest records were omitted because of missing historical climate data, and 82 nest records were omitted because one or more eggs were collected for other analyses. When multiple candidate models were constructed, models were selected based on Akaike information criterion weights (Wagenmakers & Farrell, 2004) and parsimony heuristics using the qpcR package in R (Ver 1.4-1; Spiess, 2018). Data are presented as mean ± standard error. Alpha cutoff values for significance testing (α) were set at 0.05.

Linear models and generalized linear models were constructed in R (R Foundation for Statistical Computing, 2020). Linear mixed-effects models of climate were constructed using the lme4 R package (Bates et al., 2015), and significances were determined using the lmerTest package (Kuznetsova et al., 2017). Probabilities of parasitism and nest depredation were modeled by generalized linear mixed-effects models with a fixed effect of orchard/non-orchard site and with a random year effect.

Generalized linear mixed-effects models of hatching and fledging success were built with the GLMMadaptive R package (Ver 0.7-15; Rizopoulos, 2020). Hatching success was modeled as the ratio of individuals hatched (success) to the number of destroyed or unhatched eggs (failure; Carleton et al., 2019), while fledging success was modeled as the ratio of nestlings fledged (successes) to the number of hatched but unfledged young (failure; Pinkowski, 1977). Hatching and fledging success were modeled with a beta-binomial distribution (Harrison, 2015) and a logit link function. We compared generalized mixed-effects models with fixed effects of orchard/non-orchard site, brood, and weather variables during either the incubation period (estimated to be 14 days after the first egg was laid [Gowaty & Plissner, 2020]) or the fledging period (estimated to be 18 days after the incubation period [Gowaty & Plissner, 2020]) including mean daily minimum temperature, mean daily maximum temperature, mean temperature, and total precipitation over the entire 14-day incubation period for hatching success or the 18-day fledging period for fledging success. After fledging, hatch-year birds continue to rely on parental provisioning for up to 3 weeks and disperse to new territories prior to their first breeding season; delayed dispersal, wherein a fledgling will remain in the natal territory after the hatch-year, is rare and accounts for <1% of females in cohorts with marked individuals (Plissner & Gowaty, 1996). Historical weather data for the nearest of six weather stations, all 10–15 km of the nesting sites, were downloaded from Environment and Climate Change Canada’s website using the weathercan package in R (LaZerte & Albers, 2018; Supporting Information, Table S4).

Point estimates for per-egg probability of hatching success and per-nesting probability of fledging success were obtained using population-averaged marginal coefficients of the random effects term in the GLMMAdaptive R package (Hedeker et al., 2018; Rizopoulos, 2020). Corresponding standard errors were calculated by Monte Carlo integration, also using GLMMadaptive (Rizopoulos, 2020).

Generalized linear models of hatching and fledging success as a function of orchard type, either azinphos-methyl-sprayed orchards (prior to 2012; n_{azinphos} = 1393) or modern insecticide-sprayed orchards (2012 onward; n_{modern} = 269), were constructed using the subset of 1662 nest observations made in orchards (Supporting Information, Table S1).

**RESULTS**

**Changes to phenological traits of eastern bluebirds**

We modeled the date of the first laid egg as a linear function of year, brood, and orchard (multiple R² = 0.79, p < 0.001). We found that the first egg of each brood was laid significantly earlier in the season each year over our 31-year study (Figure 1). Clutch initiation occurred 4 days earlier in the spring in 2018 compared to 1988. Site type was not a significant term (p = 0.31) in the model, which also included year (p < 0.001) and brood (p < 0.001) as significant predictors of date of first egg. The predicted dates for the first egg of the first brood were May 3, 1988, and April 30, 2018. Predicted dates of the first egg of the second brood were June 22, 1988, and June 19, 2018.

We tested the effect of site type and the effect of brood on clutch size by way of linear mixed-effects model with a random year effect. Clutch size was significantly smaller in orchards than non-orchards (estimate = −0.10 ± 0.03 eggs, t value = −2.6, p < 0.0096) and significantly smaller in the second brood than the first (estimate = −0.58 ± 0.03 eggs, t value = −17.9, p < 0.001). Mean clutch size for the first brood across all years was 4.77 ± 0.03 in orchards and 4.80 ± 0.04 in non-orchards. Mean clutch size for the second brood across all years was 4.15 ± 0.03 in orchards and 4.34 ± 0.04 in non-orchards.

We constructed linear mixed-effects models of average daily precipitation and maximum, minimum, and mean temperatures during the breeding season (May–July) as a function of year, with a random effect of the weather station where the data were collected (Table 1). Year was not a significant predictor of total precipitation during the breeding season; however, all temperature models exhibited trends of increasing daily temperature throughout the 31-year study period. We next analyzed these same weather variables taken on days when nest observations were made, modeled them as a function of site type, and found no significant differences between orchard and non-orchard sites (Table 2).
Biotic contributors to reproductive success

Next, we modeled the probability of nest parasitism as a function of orchard/non-orchard site. Site type significantly influenced the probability of nest parasitism, where orchards showed a 6.1-fold increase in probability of parasitism (Prob = 0.0075; n = 19 parasitism events, n = 1662 nest observations) over non-orchard territories (Prob = 0.0012; n = 1 parasitism event, n = 648 nest observations), although the number of nests actually parasitized was relatively small compared to the total number of nests. We observed or inferred that nest parasitism was caused by brown-headed cowbirds (Molothrus ater).

We then modeled the probability of nest depredation as a function of orchard/non-orchard site and found a significant effect of site on the probability of depredation. Orchard sites showed a 2.1-fold increase in probability of depredation (Prob = 0.065; n = 122 depredation events, n = 1662 nest observations) compared to non-orchard sites (Prob = 0.031; n = 23 depredation events; n = 648 nest observations). In the study sites, the principal author (W. F. Read) found that the most

![FIGURE 1: Julian date of first egg in the first and second broods of the breeding season for eastern bluebirds (Sialia sialis) occurs earlier each year during the 31-year study period (1988). Site type of orchard/non-orchard was not a significant predictor of clutch initiation date. The Julian date 122 corresponds to the calendar date May 2. The Julian date 173 corresponds to the calendar date June 22.](image)

| TABLE 1: Mixed-effects models of climate variables in southern Ontario as a function of year (1988–2018)* |
|-------------------------------------------------|-----------------|-----------------|-----------------|-----------------|
| Response variable                               | Estimate (per year) | Lower | Upper | Observations | p^b |
| Maximum daily temperature (°C)                  | 0.026            | 0.015 | 0.038 | 11 208        | <0.001 |
| Minimum daily temperature (°C)                  | 0.011            | 0.00041 | 0.022 | 11 222        | 0.042 |
| Mean daily temperature (°C)                     | 0.019            | 0.0083 | 0.029 | 11 192        | <0.001 |
| Total daily precipitation (cm)                  | −0.0053          | −0.019 | 0.0089 | 11 204        | 0.46 |

*Model constructed with fixed year effect and random weather station effect using climate data collected during every breeding season (May–July) for eastern bluebirds (Sialisialis) in southern Ontario from 1988 to 2018.

^bAlpha cutoff values for significance testing (α) were set at 0.05.
commonly observed nest predators were house sparrows (*Passer domesticus*), house wrens (*Troglodytes aedon*), and raccoons (*Procyon lotor*). Less commonly recorded predators were deer mice (*Peromyscus maniculatus*) and honey bees (*Apis mellifera*).

### Hatching and fledging success

To compare reproduction outcomes between orchard and non-orchard sites, we modeled the probability of successful hatching and fledging by generalized linear mixed-effects models with predictor terms of site type and brood and a random year effect (Table 3). We also ran these models excluding nests that were parasitized or depredated. Exclusion of these nests did not affect the significance of the site term for hatching or fledging success, although brood effect was not significant in the hatching success model after the exclusion of parasitized and depredated nests. Because our main finding was not different between these models, we opted to include the full data set in our analyses.

Orchard sites had significantly lower probabilities of hatching in both broods of the breeding season (Figure 2). In the first brood of the season, the probability of hatching successfully in orchard sites was 0.84 ± 0.01 and 0.88 ± 0.01 in non-orchard sites. In the second brood, the probability of hatching successfully in orchard sites was 0.82 ± 0.01 compared with 0.87 ± 0.01 in non-orchard sites (Figure 2). The second brood was 1.8% less likely to successfully hatch than the first in non-orchards and 2.3% less likely to successfully hatch in orchards.

The probability of successfully fledging a chick did not differ significantly between orchard and non-orchard sites.

### Table 3: Selection criteria and statistics for generalized linear mixed-effects models of hatching and fledging success of eastern bluebirds (1988–2018)a

| Model                      | Term          | Estimate | Lower | Upper | Observations | ϕ     |
|----------------------------|---------------|----------|-------|-------|--------------|-------|
| Hatching success (A)       | (Intercept)   | 2.21     | 1.90  | 2.52  | <0.001       |
| AlCw: 0.65                 | Site          | -0.35    | -0.55 | -0.15 | <0.001       |
| logL: ~2298                | Brood         | -0.17    | -0.33 | 0.0006 | 0.049       |
| Hatching success (B)       | (Intercept)   | 1.96     | 1.78  | 2.15  | <0.001       |
| AlCw: 0.25; logL: ~2299    | Site          | -0.35    | -0.55 | -0.15 | <0.001       |
| Hatching success (C)       | (Intercept)   | 1.72     | 0.83  | 2.61  | <0.001       |
| AlCw: 0.095                | Site          | -0.36    | -0.56 | -0.16 | <0.001       |
| logL: ~2299                | Brood         | -0.045   | -0.36 | 0.28  | 0.78         |
| Mean temperature           |               | 0.062    | -0.063 | 0.19 | 0.33         |
| Precipitation              |               | 0.0024   | -0.0014 | 0.0063 | 0.22       |
| Minimum temperature        | (Intercept)   | -0.076   | -0.21 | 0.057 | 0.26         |
| Fledging success (B)       | Site          | -0.054   | -0.25 | 0.14  | 0.59         |
| logL: ~2386                | Brood         | 0.56     | 0.40  | 0.73  | <0.001       |
| Fledging success (C)       | (Intercept)   | 1.40     | 1.20  | 1.60  | <0.001       |
| AlCw: 1.1e−23; logL: ~2408 | Site          | -0.054   | -0.25 | 0.14  | 0.59         |
| Fledging success (C)       | (Intercept)   | -0.08    | -1.94 | 0.16  | 0.096        |
| AlCw: 1                    | Site          | -0.020   | -0.22 | 0.18  | 0.85         |
| logL: ~2351                | Brood         | -0.30    | -0.57 | -0.027 | 0.031       |
| Mean temperature           |               | 0.09     | -0.066 | 0.25 | 0.25         |
| Precipitation              |               | 0.0024   | -0.0012 | 0.0059 | 0.19       |
| Minimum temperature        |               | 0.05     | -0.11 | 0.20  | 0.56         |

*All models presented include a term for random year effects.

aFrom three candidate models of hatching success (A, B, and C), we reported on model A because it had the highest Akaike information criterion weight.

bFrom candidate models of fledging success (A, B, and C), we reported on model A because it had the greatest ratio of significant terms to number of terms.

Alpha cutoff values for significance testing (α) were set at 0.05.

AlCw = Akaike information criterion weight; logL = log likelihood.
In the first brood of the season, the probability of fledging successfully was $0.75 \pm 0.02$ in orchard sites and $0.76 \pm 0.02$ in non-orchards. In the second brood, the probability of fledging successfully was $0.84 \pm 0.01$ in orchard sites compared with $0.84 \pm 0.02$ in non-orchard sites (Figure 2). The second brood was 9.0% more likely to fledge successfully than the first in orchards and 8.8% more likely to fledge successfully than the first in non-orchards.

**Azinphos-methyl insecticides in orchard sites**

We investigated the effect of the 2012 Canadian regulation of azinphos-methyl on hatching and fledging success by a generalized linear model. We included predictor terms of orchards before and after the azinphos-methyl ban and brood. Comparing reproduction endpoints in the orchards before and after the ban in 2012, we found no significant differences of hatching or fledging success (Table 4). Consistent with our global models (Table 3), brood was a significant term for both hatching success and fledging success (Table 4).

### DISCUSSION

Many land bird species in North America are suffering declines related to habitat loss, climate change, and anthropogenic disturbance (Scharlemann et al., 2004). Therefore, the quality of the remaining and alternate habitats such as agricultural areas becomes even more relevant to the survival of bird populations. To evaluate the long-term value of orchard habitats for birds in Ontario, we collected and analyzed 31 years of nesting data from eastern bluebirds using nest-boxes in conventionally sprayed orchards and non-orchards in southern Ontario to compare reproductive success between these habitats. First, we measured phenological characteristics of bluebirds in our study area from 1988 to 2018. Long-term studies of eastern bluebird nesting success have reported comparable mean dates for laying of the first brood (Bauldry...
et al., 1995; Pinkowski, 1977) to those found in our study. We determined that the first egg of each brood was laid 4 days earlier when comparing the beginning to the end of our study period.

We also found that birds had significantly smaller clutches in orchards compared to non-orchard sites. As predicted for bluebirds at this latitude, clutch size decreased from the first brood to the second brood of the season (Dhondt et al., 2002). We analyzed our longitudinal data for trends in abiotic characteristics of the landscape in our sites, which may be related to reproductive success in bluebirds, including climatic trends. We observed trends of annually increasing maximum, minimum, and average daily temperatures during the breeding season, from May to July. Previous studies have also found a strong relationship between bird phenology and advancing springtime weather conditions (Cotton, 2003; Shipley et al., 2020). Migratory birds, like the northern population of eastern bluebirds, face risks of reduced fecundity and reproductive success associated with changing climatic conditions (Shipley et al., 2020; Sillett et al., 2000). Eggs laid earlier may expose newly hatched chicks to cooler temperatures, particularly in the first clutch, which may have implications for fledging success (Shipley et al., 2020). However, there were no differences in climatic trends among our sites, and we did not observe a linear trend in daily precipitation in the breeding season during our 31-year study.

Next, we determined that hatching success of bluebirds is significantly lower (4%–5%) in orchards than in non-orchards. After hatching, birds in orchard and non-orchard sites were equally likely to fledge (75%–76% in the first brood, 84% in the second brood). Fledging success rates across broods among bluebirds have previously been reported to be 81.5% (Pinkowski, 1977) and 78.7% (Carleton & Pruett, 2011). These reports across broods agree closely with the rates of fledging success reported in the present study considering that both are higher than observed in our study for the first brood but lower than what we observed in the second brood. The differences observed between broods may relate to seasonally dynamic biotic and abiotic pressures including food supply, weather, and predation (Paquette et al., 2013; Pinkowski, 1977). Success rates in nest-boxes are comparable to those reported for natural cavities (Pinkowski, 1977).

Habitats transformed into agricultural lands generally yield lower reproductive success for grassland birds (Higgins, 1977; Rodenhouse & Best, 1983), even among species generally considered to be farmland birds (Chiron et al., 2014). We investigated several factors which may be related to lower hatching success in bluebirds. We asked whether predation and parasitism played a role in the observed difference in hatching success between site types. We determined that the probability of nest predation was 2.1-fold higher and that of nest parasitism was 6.1-fold higher in orchards compared to non-orchards. Nest-box management certainly plays an important role in protecting nesting species from predators, and differences in management practices can affect reproductive success (Barry, 1970). However, all the nest-boxes in our study were monitored and maintained by the principal author (W. F. Read) and, therefore, were not subject to interexperimenter variability. The threats of depredation and pressure from inclement weather are highest earlier in the breeding season (Pinkowski, 1977), likely contributing to the difference in fledging success observed between broods. Common nest predators depredate nests near non-productive lanes and borders within agricultural lands (Rodenhouse & Best, 1983). Vulnerability to nest parasitism is also significantly greater in fragmented agriculture habitats (Rodenhouse & Best, 1983; Vander Haegen, 2007). Furthermore, mechanical disruptions to nests from agricultural practices affect nesting birds and can reduce reproductive success (Higgins, 1977; McLaughlin & Mineau, 1995). Reproductive success in agricultural landscapes can also be impaired by destruction of nests from agricultural practices such as mowing (Kerschner & Bollinger, 1996). Those types of farm site-management effects could have been factors in these orchards; for example, farming machinery such as tractors or mowers may have contacted nest-boxes or their poles. Our long-term monitoring period from a large number of sites may have helped to minimize the effect of such contingent occurrences. In general, our observation was that when growers were operating machinery, they did not contact the nest-boxes.

The results of our 1988–2018 analysis are unique in that we were able to examine the hypothesized effect of azinphos-methyl after the insecticide was regulated. Azinphos-methyl can depress cholinesterase activity (Lari et al., 1994), but effects on reproduction have received less attention. Previous studies in orchards substantiate our finding that hatching success is the most sensitive endpoint to conditions on sprayed orchards. In a comparison of sprayed orchards with past organochlorine use versus organic orchards in Pennsylvania, fledging success and clutch sizes of American robins were not significantly different, but hatching success in one of two years of the study was significantly lower (Fluetsch & Sparling, 1994). Hatching success did not improve significantly in our orchards after the azinphos-methyl ban. This suggests that other factors in orchard management or chemicals including organophosphates, pyrethrin, and systemic and spinosad insecticides and miticides that continued to be used in orchards after 2012 (Ontario Ministry of Agriculture, Food, and Rural Affairs, 2021) may have had individual or combined toxic effects to reduce hatching success in bluebirds. Between 1993 and 1994, American robins nesting in conventionally sprayed apple orchards with past DDT use and active azinphos-methyl use in the Okanagan valley, British Columbia, Canada, had significantly lower hatching success but not fledging success compared to non-orchard sites (Gill et al., 2000). In laboratory studies, there was no significant effect of dosing with azinphos-methyl and mancozeb, either alone or in combination with p,p′-DDE on reproductive traits (timing of laying, egg size and number, yolk precursor levels) or immune status (percentage of hematocrit and leukocrit and H/L ratio) of breeding female zebra finches (Taeioppygia guttata; Gill, 2003). In contrast, p,p′-DDE in eastern bluebird eggs was significantly associated with increased occurrence of unhatched eggs in 1988–1994 in a study conducted in the same orchard and non-orchard sites as the present study in Ontario (Bishop et al., 2000).
In 2000–2001, mean p,p′-DDE concentrations ranged from 6.18 to 48.11 µg/g in eastern bluebird eggs sampled from four orchards (Mayne et al., 2004), which were also among those sampled in our study. At that time, pesticide applications in the apple orchards consisted of 16 different pesticide compounds and 10 different mixtures in 2000 and 2001 (Mayne et al., 2004). Eastern bluebird chicks from those orchards were less responsive to challenge with adrenocorticotropic hormone (ACTH), and a significant negative association was found between the response to ACTH challenge and p,p′-DDE concentration in eggs but not the current-use pesticide exposures (Mayne et al., 2004). Examination of immune organs revealed that tree swallow (Tachycineta bicolor) chicks nesting in the same study sites as the present study had significantly greater thymic lymphocyte density and cortical/medullary ratios and significant splenic B-cell hyperplasia in orchard nests relative to chicks in the non-orchard sites. Modulation of the hypothalamus–pituitary–adrenal axis in the songbird chicks tested were most associated with high p,p′-DDE egg concentrations rather than the pesticides in use at that time and in our study apple orchards in 2000–2021. Mayne et al. (2004) concluded that an alteration in the endocrine or immune system may compromise songbird survival immediately after fledging or during migration.

Although those studies were conducted in the 1990s and early 2000s, more recent data indicate that p,p′-DDE persists at elevated concentrations in birds nesting in apple orchards in Canada (Kesic et al., 2021). In apple, cherry, and pear orchards in the Okanagan valley of British Columbia, p,p′-DDE detected in American robin eggs collected in 2019 remains at relatively high concentrations (mean 36.6 µg/g, range 3.28–107 µg/g) in eggs collected from study orchards which historically applied organochlorine pesticides including DDT (Harris et al., 2000; Kesic et al., 2021). The concentrations measured in the mid-1990s in Okanagan valley orchards (approximately 2-fold higher than 2019 levels) were correlated with adverse effects on growth and brain development in American robins (Iwanuki et al., 2006), close relatives of eastern bluebirds, belonging to the same family, Turdidae, and similarly feeding on ground-dwelling invertebrates (Gowaty & Plissner, 2020; Vanderhoff et al., 2016). Overall, the evidence suggests that p,p′-DDE persistence is the most probable chemical detriment to hatching success in eastern bluebirds in our Ontario apple orchards over this 31-year period, rather than current- or recent-use pesticides. This long-term study highlights the value of examining reproduction in combination with biotic and abiotic factors that could alter reproductive success of songbirds nesting in agricultural habitats.

Supporting Information—The Supporting Information is available on the Wiley Online Library at https://doi.org/10.1002/etc.5218.

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Conflicts of Interest—The authors declare that there are no conflict of interests.

Author Contributions Statement—W.F. Read(670,514),(702,523): collection of field data. S.G. English: statistical analysis and writing of original manuscript. K. Hick: technical assistance and aggregation of field data. C.A. Bishop: study design. All authors contributed to drafts and reviewed the final manuscript.

Data Availability Statement—Data, associated metadata, and calculation tools are available from the corresponding author (christine.bishop@canada.ca).

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