





# Osmia cornuta Is a More Suitable Managed Pollinator for Cherry and Apple Orchards Than Osmia bicornis

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#### **ABSTRACT**

Managed crop pollination strongly relies on the honeybee Apis mellifera and the bumblebee Bombus terrestris, which is risky, may impact wild pollinator communities and does not always give the best pollination outcomes. The mason bees Osmia cornuta and Osmia bicornis are increasingly used as alternative crop pollinators, but it is not clear how suitable these species are for fruit pollination. Here we assessed the suitability of O. cornuta and O. bicornis as managed pollinators in two cherry and four apple orchards. O. cornuta and O. bicornis were introduced in the orchards at the start of blooming. We assessed their time till emergence, nesting rate, abundance in orchards via transect walks, flower visitation rate in 20-min time counts and pollen composition on their bodies. O. cornuta emerged  $4\pm 2$  days earlier and had a minimum of 5-fold higher nesting rate than O. bicornis in cherry and apple orchards. Transect walks revealed that O. cornuta was more abundant in orchards than O. bicornis (21.2%  $\pm$  10.1% vs.  $1.3\% \pm 1.8\%$  of all observed pollinators) and more frequently observed visiting flowers (53 vs. 5 visits for O. cornuta and O. bicornis, respectively). Pollen loads of O. cornuta contained 95% of Rosacea plants (likely apple and cherry), whereas this was 30% for O. bicornis, indicating that O. bicornis mainly foraged outside the orchards. These findings indicate that O. cornuta is a better pollinator than O. bicornis for early blooming crops, such as cherry and apple, in the relatively cool and rainy weather conditions during our study.

#### 1 | Introduction

Many fruit and vegetable crops depend on insects for pollination. Crop pollination services are valued at 153 bn € globally (Gallai et al. 2009), with 35% of all crops being dependent on animal pollination and 15% being dependent on pollination for seed production (Klein et al. 2007). Historically, pollination was provided by populations of wild pollinators, however, due to a general decline of wild pollinators combined with the increase in the area of pollination-dependant crops (Cheung 1973; Aizen and Harder 2009; Potts et al. 2010), and there is an increased need of managed pollinators to satisfy the demand of crop pollination (Sumner and Boriss 2006; Breeze et al. 2014; IPBES 2016).

The honeybee, *Apis mellifera*, and bumblebee *Bombus terrestris* are the most commonly used managed pollinators (Southwick and Southwick 1992; Watanabe 1994). However, the strong reliance on these species can be problematic as they are not always the best pollinators for specific crops (Torchio 1990; Batra 1995; Westerkamp and Gottsberger 2000). In addition, *A. mellifera* and *B. terrestris* may impact wild pollinator communities via resource competition (Thomson 2004; Paini and Roberts 2005; Henry and Rodet 2018), transmission of diseases (Graystock et al. 2013; Fürst et al. 2014; Tehel, Brown, and Paxton 2016), and outbreeding (De La Rúa, Serrano, and Galián 2002; Jensen et al. 2005). The rise of diseases and parasites in managed pollinators such as the small hive beetle or American foulbrood poses major risks to

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the economic viability of managing honey bees and bumblebees (IPBES 2016). Therefore, there is a need for a diversification of managed crop pollinators.

The use of solitary bees could diversify managed pollination services and reduce risks of crop pollination which relies currently on one or two species. Mason bees of the genus Osmia (Hymenoptera: Apidea) are promising candidates as managed crop pollinators and are used on a limited scale in North America, East Asia and Europe (Bosch, Sgolastra, and William 2008). In Europe, O. cornuta (Latreille, 1805) and O. bicornis (Linnaeus, 1758) (Hymenoptera: Megachilidae) are already used as managed pollinators in Switzerland, France, Italy and Germany (Torchio 1987; Splitt, Schulz, and Skórka 2022). Osmia bees often emerge in early spring (Westrich 1989) and forage at lower temperatures and in less favourable weather conditions than A. mellifera, therefore ensuring pollination during periods with adverse weather conditions (Vicens and Bosch 2000a). In addition, Osmia bees have a high pollination efficiency because of their pollen collection behaviour (Bosch and Blas 1994; Brittain et al. 2013; Matsumoto, Abe, and Maejima 2009) in combination with storage of collected pollen as a dry powder on their ventral abdomen. In almonds, a flower visit by O. cornuta had a 21.8%-38.1% probability of a successful pollination event, whereas this was 16.7%–25.9% for A. mellifera (Bosch and Blas 1994). Due to their limited foraging range of approximately 50-150 m (Sedivy and Dorn 2014) and a high consistency to single plant species during pollen collection, Osmia species can be applied in a very targeted way (Radmacher and Strohm 2010; Westrich 1989). Moreover, the gregarious nesting behaviour of Osmia in artificial nests allows easy breeding, transportation and mass release in crops (Cane, Griswold, and Parker 2007). Osmia bees are less likely to have a negative impact on wild pollinator communities than managed honeybees and bumblebees because they are more flower-specific, have a shorter foraging range, a shorter adult life span and their need for specific climatic and nesting conditions allow a more controlled introduction into crops (Donovan 1990). Therefore, indigenous Osmia species are good candidates to provide pollination services in early flowering, high-value crops, such as cherries, almonds or apples.

While requirements for the rearing and management of O. cornuta and O. bicornis have been relatively well studied for these species independently, their relative suitability as managed pollinators for early flowering crops is less well documented. O. bicornis needs higher temperatures for emergence than O. cornuta (Kehrberger and Holzschuh 2019), in Spain and in Serbia O. cornuta was active in rainy and windy weather conditions at 10°C-12°C, whereas O. bicornis needed temperatures above 15°C in Serbia (Vicens and Bosch 2000a; Stanisavljevic' 2000). Even though O. cornuta and O. bicornis are considered generalist, their foraging behaviours may differ substantially. For instance, the preferred host plants of O. cornuta are Sorbus species (Kratschmer et al. 2020), whereas O. bicornis prefers Salix and Acer (Hansted et al. 2014). Here we studied the suitability of O. cornuta and O. bicornis in Swiss cherry and apple orchards. Specifically, we assessed their time of emergence, nesting rate, abundance in orchards, flower visitation rate, and pollen composition on their bodies to make inferences about their host plant range. Insight into these key characteristics can help growers make informed decisions about suitable mason bee species for crop pollination.

#### 2 | Materials and Methods

#### 2.1 | Study Area

The study was conducted in two cherry and four apple orchards in north-east Switzerland (Table 1). The cherry orchard in Waldkirch (Saint-Gall) received a nesting unit for O. cornuta and O. bicornis at a mutual distance of 85 m. The second cherry orchard was located in Wädenswil (Zürich) and consisted of two smaller orchards in close proximity (1A and 1B). O. cornuta was introduced in orchard 1A and O. bicornis in orchard 1B (Table 1). The apple orchards were located in the areas of Berg (Thurgau) and Wädenswil. Due to the larger size of the apple orchards, all four orchards were equipped with one nesting unit of O. cornuta and one nesting unit of O. bicornis, which were separated by at least 100 m. Nesting units of O. cornuta and O. bicornis were assigned randomly. No other nesting unit were present in the orchards, and therefore, there were a total of 12 nesting units introduced in the six orchards of our study (six for O. cornuta and six for O. bicornis; Table 1). The type of land use in a radius of 500 m around each site was analysed via satellite imagery (Table 1).

#### 2.2 | Bees

Cocoons of *O. cornuta* and *O. bicornis* used in this study were provided by Wildbiene + Partner AG (Zürich, CH). The bees were reared in orchards in northern Switzerland during spring/summer 2018. Bees were removed from their nesting material in autumn 2018 and wintered at  $0^{\circ}\text{C} \pm 3^{\circ}\text{C}$ . The diapause of the bees was broken before their release into the study sites to ensure the synchronisation between bee emergence and orchard bloom, which is a standard procedure for commercial mason bees (Bosch and Kemp 2002). This was achieved by exposing the bees to  $10^{\circ}\text{C} \pm 3^{\circ}\text{C}$  and keeping them again at  $0^{\circ}\text{C}$  after the emergence of the first male (30 days for *O. bicornis* and 4 days for *O. cornuta*) until the beginning of bloom (see Table 1 for the exact release dates).

#### 2.3 | Nesting Sites

In each study site, one or two (Table 1) wooden nesting units were established at 1.5 m above the ground containing 500–700 *Arundo donax* stalks of 20cm length as nesting material (Appendix S1). Nesting units were placed at the centre of the orchard following the recommendations of Gruber et al. (2011) and were located at least 80 m from the edge of the orchard. Each nesting unit received 500 bee cocoons in open containers at the beginning of bloom, reflecting standard management practices (Monzón, Bosch, and Retana 2004).

## 2.4 | Comparison of Two *Osmia* Species in Their Suitability as Orchard Pollinators

Several aspects of the *Osmia* bee behaviour were monitored for each nesting site: emergence rate and time, nesting behaviour, collected pollen composition and flower visitation rate. Observations were conducted during 22 days for cherry and 18 days for apple considering the weather conditions and the blooming period of the orchards.

 TABLE 1
 Characteristics of the six study sites in north-east Switzerland.

Orchard	Location	Site	Experimentally introduced species	Type of surrounding (500 m radius)	Management practices	Introduced managed pollinators (by farmers)	Blooming period start
Cherry	Wadenswill	14	O. cornuta	70% Agricultural land, 20% residential area and 10% forest	Conventional	A. mellifera	01 April 2019
	Wadenswill	11B	O. bicornis	80% residential area, 10% agricultural land and 10% forest	Conventional	A. mellifera	01 April 2019
	Waldkirch	2	O. cornuta and O. bicornis	60% forest, 40% Agricultural land	Conventional	A. mellifera	10 April 2019
Apple	Wadenswill	8	O. cornuta and O. bicornis	60% Agricultural land, 40% residential area	Conventional	A. mellifera	15 April 2019
	Wadenswill	4	O. cornuta and O. bicornis	70% Agricultural land, 20% residential area and 10% forest	Conventional	A. mellifera	15 April 2019
	Berg	w	O. cornuta and O. bicornis	70% Agricultural land, 20% residential area and 10% forest	Organic	A. mellifera and B. terrestris	18 April 2019
	Berg	9	O. cornuta and O. bicornis	70% Agricultural land, 20% forest and 10% residential area	Organic	A. mellifera and B. terrestris	18 April 2019

#### 2.4.1 | Emergence Rate

The emergence of Osmia bees was monitored in orchards and was compared to a control group of bees that emerged under ambient indoor temperatures. In the field, the emergence rate and time of the bees was assessed every 2 days. In each nesting unit, the empty and enclosed cocoons were counted, and the empty cocoons were discarded. After 18 (apple) or 21 (cherry) days, the remaining closed cocoons were removed from the field and dissected to check for the presence of parasitoids. As the bloom of the different field sites started on different days (Table 1), the release of the bees and emergence rate assessment for the individual field sites did not start and end simultaneously.

The control group of cocoons consisted of four sets of three batches of 50 cocoons per species that were exposed to ambient indoor temperatures at four different dates to account for the fact that the emergence rate of Osmia bees accelerates during the season (e.g., bees will emerge faster in late April than early April). The first starting date was 1 April 2019 (corresponding with cherry bloom in Wädenswil), followed by 10 April (cherry bloom in Waldkirch), 15 April (apple bloom in Wädenswil) and 18 April (apple bloom in Berg). The emergence of the bees from the cocoons was checked daily and empty cocoons were removed. After a minimum of 18 days, the remaining cocoons were dissected to check for the presence of parasitoids.

#### 2.4.2 | Nesting Rate

In parallel with the emergence time observations, the establishment rate of the bees in the nesting units was monitored to assess whether the emerged bees were established in the orchard. The assessment was carried out over eight rounds at 2-day intervals during the first 10 days, beginning 2 days after the bee release, spanning a total of 18 days. Each nesting unit was observed for 20 min and the number of nesting female individuals was estimated. The females were distinguished via the reed tubes that they used for nesting. The bees are very consistent during nesting, and barely ever switch between cavities before they have finished their nest, which takes several days. Reed tubes were identified as nests of an individual, by observing the entry of the female while showing typical nesting behaviour. Each nest was then marked with a pen to avoid double counts. For each round of observations, a new colour was used to distinguish between different observation rounds. This procedure enabled the identification of tubes used as nests and the monitoring of the Osmia population in the nesting unit.

#### 2.4.3 | Pollen Composition

The host range of *O. cornuta* and *O. bicornis* was assessed by analysing the pollen load of nesting females in each nesting unit. Pollen sampling took place from the moment of emergence of the adults until the end of the blooming period. Bees were collected in front of the nest and their pollen was sampled from their abdominal brush with a toothpick. This resulted in 40 pollen samples for *O. cornuta* and 8 for *O. bicornis* in cherry orchards and 78 samples for *O. cornuta* and 41 for *O. bicornis* in apple

orchards. The pollen samples were sent for microscopy grain pollen analysis to CREAF (Ecological and Forestry Applications Research Centre, Spain). Pollen of each sample were identified at the family or genus level and counted. The pollen classified as Rosacea family was considered a proxy for pollen collected by bees on cherries and apple flowers, as there were very few other Rosaceae plants blooming near the orchards during the study period.

#### 2.4.4 | Flower Visitation

The assessment of pollinator activity in the study orchards was conducted by observation of single fruit trees. These trees located 10 m south of each nesting unit were assessed at three different blooming stages (20% of open flowers, full bloom and 20% of flowers remaining). Each observation was conducted for 20 min, and flower visits by the following pollinators were recorded: *A. mellifera*, *B. terrestris*, *O. cornuta* and *O. bicornis*. In addition, the number of flower visits and the duration of each flower visit were recorded for each observed pollinator.

#### 2.4.5 | Pollinator Survey

The abundance and diversity of pollinators in the orchard was assessed by 30-m transect walks every 2 days after the introduction of cocoons for 12 rounds for cherry and 9 rounds for apple orchards. Transects were established in the parallel fruit tree row south of each nesting unit. During transect walks of 3 min, each pollinator observed on the left tree row of the transect was identified as *A. mellifera*, *B. terrestris*, *O. cornuta* male or female, *O. bicornis* male or female, hoverfly, butterfly or other pollinator and recorded.

#### 2.5 | Data Analysis

Emergence time of cocoons (response variable) under field and ambient conditions was analysed with a Kaplan–Meier survival estimator assuming an exponential distribution with constant hazard and censoring, as not all bees emerged during the observation period. Parametric analyses were performed using the 'survreg' function of the R package 'survival' on subsets of the data in which the bee species or the type of conditions (control vs. field conditions) were selected as explanatory variables to allow a pairwise comparison by strata. Proportions of emerged and parasitised bees in each of the populations (*O. cornuta* vs. *O. bicornis*) were analysed by a Fisher exact test for count data.

Nesting rate was expressed in terms of the cumulated number of observed occupied tubes for each observation. Since assumptions for repeated measurement ANOVA were not met, we used a Wilcoxon rank-sum test with continuity correction to analyse the number of occupied tubes for each observation day for *O. cornuta* and *O. bicornis*.

Each pollen sample was categorised according to the dominant pollen type. If more than 80% of the pollen collected by a bee was from one plant type, the pollen sample was categorised as Salix, Ranuculacea, Quercus, Rosacea or other plants. Fourteen

pollen samples (<10% of samples) were not dominated by the pollen of a specific plant type and were excluded from the analysis. Excluded pollen samples represented 10% and 7% of the samples for *O. bicornis* and *O. cornuta*, respectively. A Pearson's chi-squared test was performed on the proportion of visited plant families according to the bee species (explanatory variable), followed by a Fisher exact test for count data of the visits of Rosacea for both bee species. The figure was produced with the package 'ggstatplot' (Patil 2021).

The averages of the time spent per flower per observed insect and the number of visited flowers (both response variables) were analysed by Kruskal–Wallis one-way analysis of variance for each managed pollinator (*A. mellifera*, *B. terrestris*, *O. bicornis* and *O. cornuta*). In case of significance detected by the omnibus test, a post hoc test for pairwise multiple comparisons using a pairwise Wilcoxon rank-sum test with Bonferonni correction was conducted. All the data analyses were performed with R i386 3.6.0 and Rstudio Version 1.2.1335. Means ± SEM are reported throughout the paper.

#### 3 | Results

#### 3.1 | Experimental Conditions

The 2019 blooming season for cherries started on 1 April in Wädenswil and Dietikon and 10 April for the orchard in Waldkirch. The weather conditions during the first 2weeks of April were unfavourable for bee emergence and activity, as only 5 days had temperatures exceeding 10°C without strong wind or heavy precipitation. The blooming season for apple trees started on 15 April in Wädenswil and 18 April in Berg. The weather during the two last weeks of April was more suitable for bee emergence and activity with 13 days of favourable conditions. However, a cold period in early May resulted in very much reduced activity of the bees.

#### 3.2 | Emergence Speed of O. bicornis and O. cornuta

In both cherry and apple orchards, O.cornuta emerged significantly faster than O.bicornis (Figure 1, p < 0.001 for both orchard types). In cherry orchards, at least 50% of O.cornuta emerged within 6 days after introduction (7 and 16 April for Wädenswil and for Waldkirch, respectively), whereas it took 12 and 16 days for at least 50% of all O.bicornis to emerge under the same conditions (17 and 22 April for Wädenswil and Waldkirch, respectively). In apple orchards, more than 50% of O.cornuta emerged within 4 days after introduction (19 and 23 April for Wädenswil and for Berg, respectively). O.bicornis had a slower emergence rate than O.cornuta with more than 50% emergence reached after 6 days (21 and 25 April for Wädenswil and for Berg).

Under room conditions, *O. cornuta* also emerged significantly faster than *O. bicornis* for the blooming period of cherry (1 and 10 April; p < 0.01). However, there was no significant difference between the bee species under controlled conditions during the apple blooming period (15 and 18 April). For both bee species and both orchard blooming periods, the emergence under field conditions was significantly slower than the emergence under

room conditions (Figure 1). The percentage of emergence of *O. bicornis* was significantly lower than for *O. cornuta* (76.6% vs. 87.3%; p < 0.001), whereas there was no significant difference in parasitism (1.5% of parasitised cocoons for both species, p = 0.21).

#### 3.3 | Nesting Rate of O. cornuta and O. bicornis

In apple orchards, nesting rates of  $O.\ bicornis$  were lower than for  $O.\ cornuta$  (Figure 2). From the second to 18th day after cocoon introduction, significantly more  $O.\ cornuta$  nesting bees were observed than for  $O.\ bicornis$  (p < 0.05 for each assessed day). In cherry orchards, the first  $O.\ bicornis$  nesting behaviours were observed on 16 and 12 days after introduction in Wädenswil and Waldkirch (17 and 22 April, respectively), whereas  $O.\ cornuta$  nesting commenced on day 8 in both locations (9 and 18 April, respectively). The number nests were too low for a meaningful analysis of the comparison of nesting behaviour of  $O.\ bicornis$  and  $O.\ cornuta$ .

#### 3.4 | Hostplant Range

The composition of the pollen on the bodies of *O. cornuta* and *O. bicornis* was different for both orchards. In cherry orchards, 80% of the *O. cornuta* individuals (n=40) had a pollen composition with more than 80% Rosacea pollen, whereas 13% of the pollen samples from *O. bicornis* (n=8) contained Rosacea pollen. The sample size from *O. bicornis* in cherry orchards was too low for a meaningful statistical analysis. In apple orchards, *O. cornuta* (n=78) and *O. bicornis* (n=37) pollen samples contained 95% and 30% of Rosacea pollen, respectively, which was significantly different (p<0.001; Figure 3).

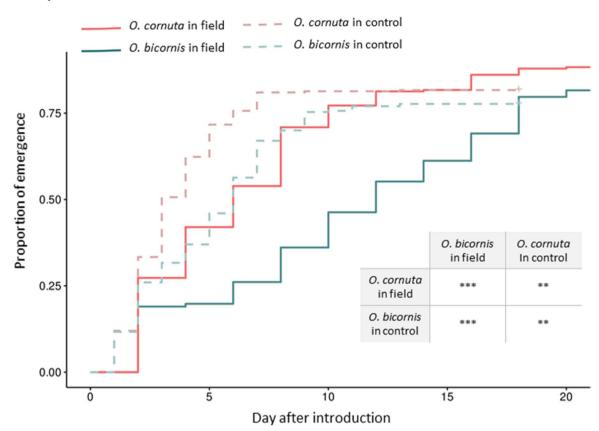
#### 3.5 | Flower Visitation and Pollinator Surveys

In cherry orchards, 69 *O. cornuta* and 31 *A. mellifera* individuals were recorded during the observation period. The average time spent per flower of  $5.52 \pm 2.52 \, \mathrm{s}$  and  $12.29 \pm 7.01 \, \mathrm{s}$ , respectively (p < 0.001). The number of flowers visited per individual was not significant between species ( $3.97 \pm 0.54$  flower visited/tree/individual for *A. mellifera* and  $4.38 \pm 0.40$  for *O. cornuta*). No flower visitation was recorded for *O. bicornis*.

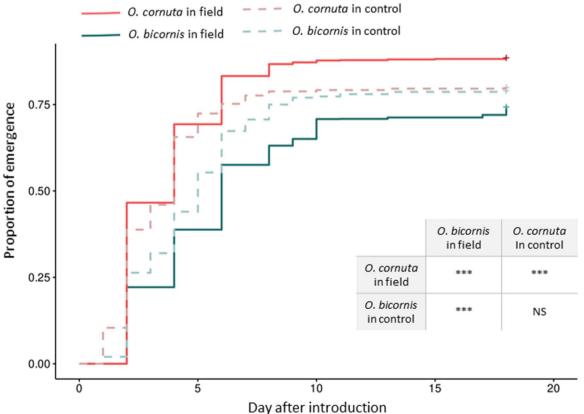
In apple orchards, four different pollinators were observed: *A. mellifera* (n=66), *B. terrestris* (n=29), *O. bicornis* (n=5) and *O. cornuta* (n=53). *A. mellifera* took significantly more time when visiting flowers than *B. terrestis*, *O. bicornis* and *O. cornuta* (respectively p<0.001, p=0.004 and p<0.001; Figure 4). However, the average number of flowers visited per tree was not significantly different between species ( $5.91\pm0.59$  flower visited/tree/individual for *A. mellifera*,  $6.21\pm0.95$  for *B. terrestis*,  $4.00\pm1.64$  for *O. bicornis* and  $4.85\pm0.50$  for *O. cornuta*).

The pollinator community (n = 945) during blooming periods of cherry and apple was dominated by managed species ( $91.84\% \pm 5.86\%$ ) with  $62.23\% \pm 17.84\%$  *A. mellifera*,  $21.18\% \pm 10.08\%$  *O. cornuta*,  $7.18\% \pm 7.57\%$  *B. terrestris* and  $1.25\% \pm 1.84\%$  *O. bicornis*.

### A. Cherry





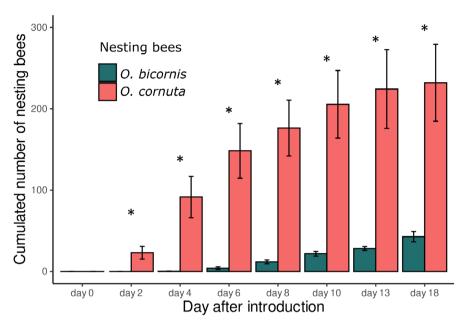


 $\textbf{FIGURE 1} \quad | \quad \text{Legend on next page}.$ 

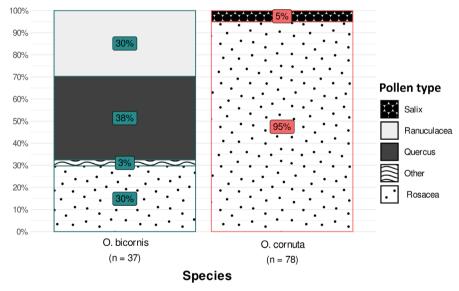
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**FIGURE 1** | Proportion of emerged *O. cornuta* and *O. bicornis* cocoons over time after introduction at the beginning of the blooming period in cherry orchards (A) and apple orchards (B). Field conditions are in solid lines, controlled conditions are in dashed lines, *O. cornuta* emergence are in red lines and *O. bicornis* emergence are in blue lines. Tables show significance levels of Kaplan–Meier parametric analyses on subset data for pairwise comparison. \*\*\*\*p < 0.001, \*\*p < 0.01, NS, not significant. [Colour figure can be viewed at wileyonlinelibrary.com]



**FIGURE 2** | Cumulative number of occupied nesting cavities by *O. bicornis* and *O. cornuta* in nesting sites in apple orchards over the blooming period. From day 2 to day 18, *O. cornuta* occupied significantly more nesting cavities in nesting sites. \*p < 0.05. [Colour figure can be viewed at wileyonlinelibrary.com]

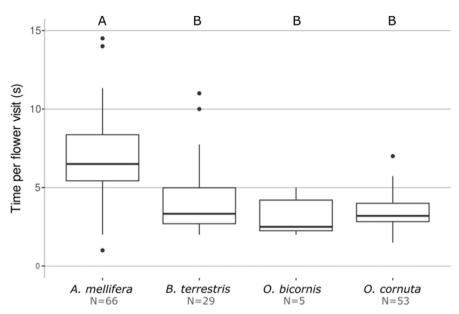


**FIGURE 3** | Proportions of plant families/genera visited by *O. bicornis* and *O. cornuta* for pollen collection in apple orchards. When more than 80% of pollen collected by an individual was from the same plant species group, it was counted as a visit for this group (Fisher exact test: p < 0.001). [Colour figure can be viewed at wileyonlinelibrary.com]

#### 4 | Discussion

Crop pollination depends heavily on the use of managed honeybees and bumble bees. Osmia bees may be suitable candidates to diversify the managed pollination in fruit orchards (Bosch and Blas 1994; Eeraerts et al. 2020), but the relative suitability of the species in this genus is still poorly documented. Here we assessed the suitability of *O. cornuta* and

O. bicornis in cherry and apple orchards of Switzerland. We found that O. cornuta is a better pollinator in both apple and cherry orchards as it emerges and nests faster and in greater numbers than O. bicornis. Moreover, the collected pollen samples indicate that O. bicornis visits a much wider range of host plants and is therefore an unreliable pollinator compared to O. cornuta, which seems to visit mainly the target orchard crops.



**FIGURE 4** Average pollinator flower visitation duration on apple flowers by four different managed pollinators. 'N' indicates the number of observations per bee species. p < 0.001 for Kruskal–Wallis rank-sum test, letters indicate a p value of < 0.01 for pairwise comparisons using Wilcoxon rank-sum test with Bonferroni correction.

We found that O. cornuta had more favourable characteristics than O. bicornis for the pollination of cherry trees. Cherry is a challenging crop for pollination in Switzerland because of its early bloom at the end of March and early April, when weather conditions can be unfavourable for pollinators. The emergence rate of O. bicornis was low and pollen collection and nest building only started 2 weeks after blooming start. This was problematic for cherry production because flower visitation during the first days of flowering are regarded as critical for a good fruit set in cherries (Sanzol and Herrero 2001). In contrast, O. cornuta emerged considerably faster, allowing them to nest and pollinate in the second week of cherry bloom. This finding corresponds with reports that O. bicornis needs significantly higher temperatures for emergence than O. cornuta (Kehrberger and Holzschuh 2019). It was shown that O. cornuta increases rapidly in supercooling points, indicating a more abrupt diapause completion than O. bicornis, which exhibits a slower, more gradual rise (Krunić and Stanisavljević 2006). This aligns well with the behaviour of natural populations of O. cornuta and O. bicornis. O. cornuta is much earlier active in spring, for example, from Mid-March to Early May in Switzerland and Southern Germany, whereas O. bicornis appears much later in the year, for example, from Early April to Mid-June (Westrich 1989). The activity threshold of O. cornuta is approximately 9.8°C, and lower than that of A. mellifera at 12.5°C (Vicens and Bosch 2000b). O. bicornis has comparable or lower activity thresholds than A. mellifera (Hansted et al. 2015), indicating a lower threshold compared to O. cornuta. All this information indicates that O. bicornis emerges later in spring than O. cornuta and has also a higher associated temperature threshold for activity and nesting.

Under the unfavourable weather conditions during our study, *O. cornuta* probably also performed better than honeybees, which have an activity threshold of 12.5°C (Vicens and Bosch, 2000a), which exceeds the temperatures of most days during our study. In addition, the few samples of pollen collected by *O. bicornis* suggest that *O. bicornis* mostly visited *Ranunculaceae* and

Quercus and only rarely cherry trees. While care is needed for the interpretation of these data because of the small sample size (n=8), other studies confirm that pollen collected by *O. bicornis* that were released in fruit orchards are often dominated by non-Rosaceae pollen, such as Quercus, Salix or Acer pollen (Ruddle et al. 2018; Hansted et al. 2014). The far majority of pollen on the bodies of *O. cornuta* originated from Rosacea plants, indicating that they mostly visited the target crop. The pollen composition recorded for *O. cornuta* is in line with earlier studies that show a strong preference of this species for cherry and almond (Márquez, Bosch, and Vicens 1994). Based on the evaluation on the above characteristics, we consider *O. cornuta* a more suitable pollinator for cherry than *O. bicornis*.

In apple orchards, which bloom approximately 2 weeks later than cherry, we also found that O. cornuta emerged faster and started nesting and pollen collection earlier than O. bicornis. Pollination is less critical in apple than in cherry because weather conditions are usually better and growers apply fruit thinning to reduce the number of apple fruitlets to warrant a good fruit size at harvest (Byers and Carbaugh 2002) and avoid alternate bearing (Dennis and Neilsen 1999). Nevertheless, even under these more favourable conditions, we observed a clear difference between O. cornuta and O. bicornis. For instance, relatively few O. bicornis females were active around the nesting sites compared to O. cornuta, which may indicate a higher dispersal rate of O. bicornis compared to O. cornuta. Dispersal events have been observed in mason bees after disturbance or due to unfavourable nesting conditions (Vicens and Bosch, 2000b; Gruber et al. 2011), with a certain degree of dispersal considered normal (Gruber et al. 2011). As few or no alternative nesting sites were available in our orchards, this suggests that O. bicornis left the orchards to settle somewhere else. However, as there are no comparative studies regarding the cause and extend of dispersal behaviour between Osmia species, the observed differences could also be explained by other factors, such as an increased mortality of females.

The relatively high nesting rate of *O. cornuta* most likely resulted in a sufficient population for a satisfactory pollination of apple orchards, even though we did not assess this, while this was not the case for *O. bicornis*. While the early-season emergence and nesting behaviour of *O. cornuta* have been reported in Spain and Serbia (Bosch 1994; Krunić and Stanisavljević 2006), this has not yet been observed in central Europe.

O. cornuta pollen loads were dominated in 95% of all samples by pollen from Rosacea plants, indicating a clear preference for apple flowers. In contrast, pollen loads of O. bicornis were only dominated in 30% of all samples by pollen from Rosacea plants, with Quercus pollen being very prevalent (38% of samples). It is well established that O. bicornis prefers non-crops plants over crop plants (Hansted et al. 2014; Márquez, Bosch, and Vicens 1994; Ruddle et al. 2018) and that O. cornuta prefers fruit trees (Márquez, Bosch, and Vicens 1994; Monzón, Bosch, and Retana 2004). The preference of O. bicornis for Quercus pollen may be due to its capacity to speed up early-season nest construction (Persson, Mazier, and Smith 2018) and enhance reproductive success (Yourstone et al. 2021), but the reason for the preference for Quercus pollen remains to be elucidated (Eckerter et al. 2022). The foraging behaviour of O. cornuta is therefore more suitable for apple and cherry pollination than the more diversified collection behaviour of O. bicornis.

In 80 transect walks in cherry and apple orchards, we only found 76 wild pollinators, indicating that crop pollination strongly depends on managed pollinators, which included mason bees, honeybees and bumblebees. With the early blooming periods and unpredictable weather conditions, the visits of non-managed pollinators can presumably not provide enough pollination for satisfactory fruit set in commercial orchards (Rader et al. 2009). In our study, the majority of observed pollinators were honeybees, which can be explained by the high honeybee hive density around our orchards (Table 1). However, flower visitation by honeybees alone may not provide sufficient pollination as the observed 2.5 visits/tree/20 min for *A. mellifera* in our orchards was far below the recommendation of 20 honeybee visits/tree/min (FAO 2018). This underlines that there is merit for the use of other managed pollinators, such as *O. cornuta*.

Despite the clear differences between the emergence rates of *O. cornuta* and *O. bicornis* in the field conditions and in room conditions for cherry bloom, the situation was less clear during apple bloom. Under controlled conditions, both species had comparable emergence rates during apple bloom, whereas under field conditions, there was still a strong difference observable between species, with *O. cornuta* emerging much faster, indicating that temperatures in the field were not high enough to induce emergence quickly.

The slower emergence of *Osmia bicornis* in orchards is likely due to physiological factors, influenced by wintering conditions and the requirement for higher temperatures to break diapause. *Osmia cornuta* showed a 10% higher overall emergence than *O. bicornis*, which could result from higher winter mortality in *O. bicornis* (Torchio 1987) or from undetected pathogens, though parasitoid impact was similar in both species (1.5%). There are extensive studies regarding wintering mortality of Osmia bees. The main drivers of mortality are unfavourable temperature conditions

during and before diapause initiation of the bees. Especially too long or too short exposure to cold temperatures can be a major source of mortality, especially due to excessive consumption of energy reserves (Bosch and Kemp 2004; Bosch, Sgolastra, and William 2008; Sgolastra et al. 2011). Under controlled conditions, *O. bicornis* emerged more quickly and in greater numbers than in orchards, whereas *O. cornuta* showed lower emergence at room temperature than in field settings. Although the distributions of *O. bicornis* and *O. cornuta* in Europe overlap to a great extent, *O. bicornis* also occurs in northern Europe. Therefore, *O. bicornis* may require a higher temperature to break diapause than *O. cornuta* to prevent its population to suffer from fluctuating spring conditions in northern Europe as an early emergence can be detrimental when whether conditions can become hostile again.

There were a few limitations in the design of our study. First, since it was conducted during 1 year, care is needed to generalise our findings. The experiment took place during a cold, wet spring. In early April, only 5 days had favourable conditions for bee emergence (temperature > 10°C, no strong wind and no heavy rain). Conditions improved in late April, with 13 favourable days for bee activity and pollination. However, a climatic depression in early May brought frequent rain and low temperatures, limiting bee foraging and slowing down metabolism. These hostile spring conditions are mainly found in the northern and eastern part of Europe (Shongwe et al. 2007). Thus, our comparison of the two Osmia species are not representative for typical milder weather conditions in southern Europe, where O. bicornis may perform better. However, the suitability of O. bicornis as a managed pollinator for northern Europe is not conclusive. In Denmark, O. bicornis is considered less effective than A. mellifera due to its non-crop host plant range (Hansted et al. 2014), whereas in the UK and Germany, O. bicornis is considered a suitable pollinator for orchards (Ryder et al. 2020; Gruber et al. 2011). These contrasting reports may point to the capacity of O. bicornis to adapt to local conditions, whereby populations may differ in favourable traits for orchard pollination. Second, due to adverse weather conditions only very few observations of O. bicornis in cherry orchards were made and therefore not all aspects of their pollination and nesting behaviour could be assessed. Third, for the pollen composition analysis, it was not possible to distinguish the origin of the pollen to the species level for Roasacea and could therefore not distinguish between cherry, apple and other Rosacea. However, since there were mainly Rosacea plants blooming close to the orchards and only few Prunus spinosa, we consider it as a safe assumption that pollen was obtained from fruit trees and not of other trees. As the foraging range of both species is only 50-150 m (Sedivy and Dorn 2014), we can be confident that Rosacea pollen was almost exclusively collected on cherry or apple trees. Although these limitations might limit the generalisation of our results to other agroecosystems, we could still produce a strong case study for the advantages of *O. cornuta* over *O. bicornis* for the pollination of fruit orchards in continental climatic zone in Europe.

#### 5 | Conclusion

We showed that there are few wild pollinators in Swiss cherry and pear orchards, underlining the critical importance of managed pollinators to achieve satisfactory pollination. *O. cornuta*  emerged faster and started nesting earlier and in greater numbers than *O. bicornis*. This was especially the case in cherry orchards but was also observed in apple orchards. Moreover, *O. cornuta* mainly collected on cherry and apple trees, whereas *O. bicornis* often collected pollen from other plant species. This indicates that *O. cornuta* has a higher potential for commercial pollination than *O. bicornis* for cherry and apple orchards in the central part of Europe. However, *O. bicornis* may still be suitable in specific northern regions were *O. cornuta* does not naturally occur, such as in the UK. Our study supports the relevance of the introduction of managed *O. cornuta* in Swiss apple and cherry orchards to provide sustainable pollination services, even in harsh weather conditions.

#### **Author Contributions**

Laurie Magnin: conceptualization, investigation, writing – original draft, methodology, visualization, writing – review and editing, software, data curation, formal analysis. Felix Bianchi: conceptualization, methodology, writing – review and editing, supervision, validation. Steffen Hagenbucher: funding acquisition, conceptualization, writing – review and editing, validation, project administration, supervision, resources.

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#### **Conflicts of Interest**

The authors declare no conflicts of interest.

#### **Data Availability Statement**

All the data of this project and script used to analyse them are available on: LaurieMagnin/Assessment-of-two-Osmia-species-as-a-sustainable-alternative-to-Apis-mellifera-pollination-services(github.com).

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#### **Supporting Information**

Additional supporting information can be found online in the Supporting Information section.