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Abstract

abstract text

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1 Introduction

2 Methods

2.1 Study system

Our study took place in the Lower Fraser Valley in southwestern British Columbia, Canada, in an agricultural system dominated by mixed vegetable, hay, and perennial berry production. From 1984-2018 the Lower Fraser Valley underwent a 13% decrease in forest patch area, mainly resulting from conversion to urban or agricultural land use (Paul et al., 2020). .... *decide what else needs to be said in this section later on!*

Field surveys were conducted across six replicate landscapes distributed throughout the Lower Fraser Valley. Each landscape encompassed roughly 3 sq km of farmland interspersed with rural/suburban residence. Landscapes were initially chosen to span a gradient of configurational and composition diversity metrics, including Shannon's diversity, edge density, and the ratio of annual to perennial crop cultivation.

Thirty sampling transects (50 meters x 2 meters) were established in each landscape, spaced as evenly as possible based on land-access and the availability of foraging resources on which to observe bees. We did not survey in active crop fields except for high-bush blueberry, which offers floral resources during its spring bloom and can sometimes host other flowers such as clovers (*Trifolium spp.*) and flatweed (*Hypochaeris radicata*) later in

the season. A total of *INSERT TOTAL TRANSECT NUMBER HERE* were surveyed over the course of two years (2022-2023) due to changes in land access within and between years.

2.2 *Bombus* collections and floral surveys

Each landscape was surveyed during 10 sampling rounds in year one (May-August 2022) and 17 sampling rounds in year two (March-August 2023). During each round of sampling, surveys were conducted on *INSERT MEAN PLUS OR MINUS SD OF SAMPLING EFFORT* (mean $\pm$ SE) transects.

Bombus surveys at each transect entailed 5 minutes of active search time (totaling 140 hours in 2022 and *INSERT TOTAL SAMPLING EFFORT 2023* in 2023), during which the stopwatch was paused whenever a foraging bumble bee was sighted. Specimens were captured by netting, placed into sterile 15 mL tubes, and immediately placed on ice before transfer to a -80°C freezer at the end of the day. Surveys were conducted on days when the temperature was above 12°C (10°C for queen surveys) and wind speeds below 2.5 m/s. In 2022, all *Bombus* species were collected; in 2023 only the focal species (*B. mixtus* and *B. impatiens* were collected.

To assess floral quality, all flowering plants within the transect area were identified to species or genus level. Abundance estimates were taken for each species on the log-scale (i.e., 0 = 1-10 inflorescences, 1 = 11-100 inflorescences, 2 = 101-1000 inflorescences, 3 = 1001-10,000 inflorescences, 4

= 10,000+ inflorescences). Floral survey data was later filtered to exclude species which bumble bees were never observed visiting (based on over 3,400 visitation events in 2022, and 3,500 visitation events in 2023). This filtering step was included to reduce the noise introduced by a variety of herbaceous weeds with flowers too small to attract or support bumble bee foragers, but which were frequently observed on the transects in high abundance.

2.3 Landscape characterization

Land cover maps were developed for each study site based on manual classification of Google Earth satellite imagery (2021) and site visits. Briefly, land cover was classified into 16 categories: annual row crops, blueberry, cranberry, other perennials, polyculture, hay meadows, pasture, fallow, grassy field margins, hedgerows (tree-dominated), hedgerows (blackberry-dominated), forest, wetlands, urban/suburban, roads, and water). These land cover types were chosen based on their hypothesized provisioning of nesting/floral resources and differences in disturbance regimes (see Table **??** for details). Land cover was mapped at 2-meter resolution in QGIS (QGIS Development Team, 2024).

INSERT EXPLANATION OF LANDSCAPE METRICS AND HOW WE CALCULATED THEM

Table 1: Description of land cover classifications, including hypothesized resource provisioning for bumble bees. Based on personal observations and expert opinion.

Landcover Class	Description	Vegetation Level	Flowers	Nesting	Disturbance Rank
annual	monoculture annual crops, tilled yearly	bare	sometimes (resource pulse)	no	5
polyculture	mixed annual crops, tilled yearly	bare	sometimes	no	5
hay	multiple cuts per year, tilled every 1-7 yrs	grass	sometimes (clover)	some species	4
fallow	fields taken out of production temporarily	grass	yes	some species	2
pasture	grazed hay meadow	grass	yes	some species	4
blueberry	described in text	woody	yes (resource pulse)	some species	3
cranberry	perennial, flooded yearly	herbaceous	yes (resource pulse)	no	5
other perennial	orchards, tree farms, etc.	woody	sometimes	yes	3
hedgerow (blackberry)	field margins dominated by *Rubus armeniacus* and *R. laciniatus*	woody	yes (resource pulse)	yes	2
forest	forest fragments (primarily native species)	woody	yes	yes	1
hedgerow (woody)	planted or remnant hedgerows dominated by trees; planted hedgerows contain flowering species selected for pollinators	woody	yes	yes	2
road/ industrial	paved/impermeable surfaces	bare	no	no	5
grassy margins	unmanaged field margins without woody vegetation	grass	yes	some species	3
suburban/ urban	residential properties, including gardens	woody	yes	yes	4
water	lakes, rivers, irrigation ditches, ocean	bare	no	no	5

2.4 Colony assignments

We used microsatellite genotyping to assign workers (2022 and 2023) and queens (2023) to their natal colonies. DNA was extracted from the mid-leg basitarsus and tarsus (distal tarsus only for queens) using the HOT-SHOT protocol (**INSERT TRUITT**). We utilized the following microsatellite loci from the existing literature: BT10, BTERN01, BL13, BL15, B126, BTMS0057, BTMS0059, BTMS0062, BTMS0083 (both species), BTMS0066, BTMS0072, BTMS0086, BTMS0104, BTMS0126, BTMS0136 (*B. mixtus* only), BT28, BT30, B10, B96, B124, BTMS0073, and BTMS0081 (*B. impatiens* only) (**CITE STOLLE**; **REBER-FUNK**; **ESTOUP–CHECK WHICH ESTOUP PAPERS!**). One primer for each locus was individually dye-labelled using 6FAM, NED, PET, or VIC, and loci were amplified in two multiplex reactions per species. Each multiplex reaction contained 4μL of template DNA, 1μL of 10X primer mix, and 5μL of Qiagen 2X Multiplex PCR Master Mix (Qiagen, Hilden, Germany). See **??** for plexes and primer concentrations. Diluted PCR products were submitted for ...

Scoring error rates were assessed by re-genotyping a panel of 96 individuals per species, and loci with observed error rates $\geq$3% were discarded from further analyses (BL15 in both species). BTMS0072 was also discarded for *B. mixtus* due to poor amplification and difficulty assigning peak-calls.

3 Results

4 Discussion

References

Paul, S. S., Dowell, L., Coops, N. C., Johnson, M. S., Krzic, M., Geesing, D., & Smukler, S. M. (2020). Tracking changes in soil organic carbon across the heterogeneous agricultural landscape of the Lower Fraser Valley of British Columbia. *The Science of the Total Environment*, *732*, 138994. https://doi.org/10.1016/j.scitotenv.2020.138994

QGIS Development Team. (2024). *QGIS geographic information system* (manual). Open Source Geospatial Foundation. http://qgis.org

Supporting Information