

Insect pollinator monitoring in and around a netted plot of apple trees – biosafety implications for genetically engineered fruit trees

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Supplementary Materials 1: Tables and Figures

Table S1. Average temperature and total precipitation. Values during the sampling periods in 2016 to 2019 are presented based on daily data from the weather station REH, Reckenholz, Zurich, Switzerland (47.4277°N, 8.5179°E, 443m above sea level).

Sampling period	Temperature [°C]	Precipitation [mm]
9.5.-24.6.2016	15.2	320
4.4.-15.5.2017	9.8	137
6.4.-7.5.2018	14.0	6
5.4.-22.5.2019	9.7	145

Table S2. Yearly abundance of different arthropod taxa. Arthropods were collected in coloured pan traps placed inside and outside a netted plot of apple trees in Zurich, Switzerland, during the flowering period of the ‘Ladina’ and ‘Nicogreen’ trees included in the experiment in the years 2016-2019. Collected specimens of holometabolic groups were adults unless stated otherwise.

	2016		2017		2018		2019		Total	
	inside	outside	inside	outside	inside	outside	inside	outside	inside	outside
Diptera total	2010	4122	2260	4814	696	884	2611	2609	7577	12429
Asilidae	0	32	168	767	12	49	65	301	245	1149
Chloropidae	24	875	0	21	0	0	0	2	24	898
Syrphidae	0	13	0	5	0	2	0	5	0	25
other Brachycera	1877	3074	1355	2726	463	558	1979	1607	5674	7965
Tipulidae	6	3	2	5	11	0	75	1	94	9
other Nematocera	103	125	735	1290	210	275	492	693	1540	2383
Hymenoptera total	133	409	166	342	150	735	465	669	914	2155
Symphyta	0	5	5	17	10	18	33	18	48	58
parasitoids	105	107	79	76	36	60	63	97	283	340
Formicidae	11	29	10	30	11	32	12	48	44	139
<i>Bombus</i> spp.	0	4	0	5	0	3	0	0	0	12
other Apoidea	16	261	72	204	91	615	353	500	532	1580
other Hymenoptera	1	3	0	10	2	7	4	6	7	26
Coleoptera total	83	407	150	469	294	325	338	748	865	1949
Nitidulidae	41	223	108	302	272	242	273	519	694	1286
Staphylinidae	16	57	18	104	10	38	20	118	64	317
Curculionidae	1	9	3	2	1	2	2	30	7	43
Alticini	2	4	2	2	0	6	6	11	10	23
Coccinellidae	0	11	2	1	0	5	2	3	4	20
- larvae	0	0	0	1	0	0	0	0	0	1
other Coleoptera	23	103	17	57	11	32	35	67	86	259
Hemiptera total	168	228	47	126	18	39	20	63	253	456
Aphididae	145	208	34	107	17	31	16	41	212	387
Heteroptera	11	10	5	4	0	4	3	14	19	32
Cicadellidae	9	5	7	12	1	3	0	5	17	25
Psyllidae	3	5	1	3	0	1	1	3	5	12
Collembola	1209	48	31	24	33	22	3	15	1276	109
Thysanoptera	85	154	86	148	42	34	167	430	380	766
Araneae	15	15	18	8	18	9	21	21	72	53
Lepidoptera total	0	3	2	6	1	6	3	3	6	18
- adult	0	3	2	6	1	2	2	3	5	14
- larva	0	0	0	0	0	4	1	0	1	4
Neuroptera total	2	5	1	2	0	0	0	0	3	7
- adult	2	5	0	2	0	0	0	0	2	7
- larva	0	0	1	0	0	0	0	0	1	0
Psocoptera	0	0	2	1	0	0	1	1	3	2
Mecoptera	0	0	1	1	0	1	0	0	1	2
Myriapoda	0	0	0	0	0	0	0	2	0	2
Orthoptera	0	0	0	0	0	0	0	1	0	1
Acarina	0	0	0	0	0	1	0	0	0	1
Plecoptera	0	0	0	0	0	1	0	0	0	1
Dermaptera	0	0	0	1	0	0	0	0	0	1
Undetermined	0	0	0	0	0	1	0	1	0	2
Total	3705	5391	2764	5942	1252	2058	3629	4563	11350	17954

Table S3. Yearly abundance of bee species. Bees were collected in coloured pan traps placed inside and outside a netted plot of apple trees in Zurich, Switzerland, during the flowering period of the ‘Ladina’ and ‘Nicogreen’ trees included in the experiment in the years 2016-2019. The last 2 columns contain species collected before the hail net was closed in 2019. All remaining data are from collections after the net was closed. Abbreviations of species names (used for ordination plots, Figure 4 in the main manuscript and Figure S3) are composed of the first 3 letters of the genus and the species name, followed by the sex (m = male, f = female, w = worker for *Apis mellifera* and *Bombus* spp.).

Species	Sex	Abbreviation	Thorax width [mm]	2016		2017		2018		2019		Total		Open net 2019	
				inside	outside	inside	outside	inside	outside	inside	outside	inside	outside	inside	outside
<i>Andrena alfkenella</i> Perkins 1914	female	-												1	
<i>Andrena bicolor</i> Fabricius 1775	female	Andbicf	2.8			1	4	2	3	1	3	4	10	13	7
<i>Andrena bucephala</i> Stephens 1846	female	-	3.2				1						1		
<i>Andrena chrysosceles</i> (Kirby 1802)	male	-	2.0						1				1		
<i>Andrena cineraria</i> (Linnaeus 1758)	female	Andcinf	3.8		1	1	3	1			2	2	6	2	
	male	-	3.1				1		3				4		12
<i>Andrena dorsata</i> (Kirby 1802)	female	-	3.0			1				1		2		1	2
<i>Andrena flavipes</i> Panzer 1799	female	-	3.3						3				3	1	1
	male	Andflam	2.7							5	1	5	1	5	
<i>Andrena fulva</i> (Müller 1766)	female	-	4.1						1	1	1	1	2		
<i>Andrena fulvago</i> (Christ 1791)	male	-						1				1			
<i>Andrena fulvata</i> Stoeckert 1930	female	-	2.9						1		1		2	1	2
	male	-	2.6											1	
<i>Andrena grvida</i> Imhoff 1832	female	-	3.8				1				1		2		
<i>Andrena haemorrhoa</i> (Fabricius 1781)	female	Andhaef	3.6		4	1	3		2		3	1	12	2	
	male	Andhaem	2.8			1	3	2	7	1	1	4	11		
<i>Andrena helvola</i> (Linnaeus 1758)	female	-	3.1				1						1	1	
<i>Andrena humilis</i> Imhoff 1832	female	Andhumf	3.3	1	15						1	1	16	11	8
	male	-	3.0											2	2
<i>Andrena minutula</i> (Kirby 1802)	female	Andminf	2.1		4	4	11	1	1	11	4	16	20	1	3
<i>Andrena mitis</i> Schmiedeknecht 1883	female	-	3.1		1		1						2		
	male	-	2.5								1		1		
<i>Andrena nigroolivacea</i> Dours 1873	female	-	3.5		1								1		
<i>Andrena nitida</i> (Müller 1776)	female	-	4.3				1		1		1		3		1
	male	-	3.3						1				1		2
<i>Andrena ovatula</i> (Kirby 1802)	female	-	2.9						1		1		2		
	male	-	2.4							2		2			
<i>Andrena subopaca</i> Nylander 1848	female	Andsubf				4		2				6			
<i>Andrena vaga</i> Panzer 1799	female	Andvagf	4.1			1	37		14	27	78	28	129	23	21
	male	Andvagm	3.2				13		22	25	46	25	81	107	199

<i>Apis mellifera</i> Linnaeus 1758	worker	Apimelw	4.2	1	96		27		1		8	1	132	3	3
<i>Bombus pascuorum</i> (Scopoli 1763)	female	-	6.7				1						1		
<i>Bombus pratorum</i> (Linnaeus 1761)	worker	-	4.8		1								1		
<i>Bombus sylvarum</i> (Linnaeus 1761)	female	-	6.7				1		2				3		
	worker	-	4.7		1								1		
<i>Bombus terrestris</i> (Linnaeus 1758)	female	-	8.3				2						2		
<i>Bombus terrestris</i> -group	worker	-	5.5		2				1				3		
<i>Bombus veteranus</i> (Fabricius 1793)	female	-	6.7				1						1		
<i>Chelostoma florissomne</i> (Linnaeus 1758)	female	Cheflef	2.4	4						3		7			
	male	Cheflom	2.4	4		1	1	11		4	1	20	2		
<i>Colletes cunicularius</i> (Linnaeus 1761)	female	Colcunf	4.8		5		14		11	3	20	3	50	7	7
	male	Colcunm	4.2					2	384	14	13	16	397	82	113
<i>Halictus scabiosae</i> (Rossi 1790)	female	Halscaf	3.6		15		2				3		20		
<i>Halictus simplex</i> Blüthgen 1923 - group	female	Halsimf	3.0	1	39	2	5		1	4		7	45		1
<i>Halictus tumulorum</i> (Linnaeus 1758)	female	Haltumf	2.1	1	5	5	8	1	6	2	1	9	20		
<i>Lasioglossum albipes</i> (Fabricius 1781)	female	-	2.2		1								1		
<i>Lasioglossum calceatum</i> (Scopoli 1763)	female	Lascalf	2.7		11	5	8	8	55	9	43	22	117		7
<i>Lasioglossum fulvicorne</i> (Kirby 1802)	female	-	2.2				1						1		
<i>Lasioglossum glabriusculum</i> (Morawitz 1872)	female	Lasglaf	1.4	1	4	5	7				10	6	21		
<i>Lasioglossum laticeps</i> (Schenck 1868)	female	Laslatf	2.2	1		2	2	2	7	9	9	14	18		
<i>Lasioglossum lativentre</i> (Schenck 1853)	female	-	2.3		1								1		
<i>Lasioglossum leucozonium</i> (Schränk 1781)	female	Lasleuf	2.7		10								10		
<i>Lasioglossum malachurum</i> (Kirby 1802)	female	Lasmalf	2.5		33	22	30	53	63	203	155	278	281	160	174
<i>Lasioglossum morio</i> (Fabricius 1793)	female	Lasmorf	1.6				1			2	4	2	5		
<i>Lasioglossum pallens</i> (Brullé 1832)	female	-	2.5		1	1						1	1		
	male	-						1				1			
<i>Lasioglossum pauxillum</i> (Schenck 1853)	female	Laspauf	1.8	2	5	15	5	3	2	21	11	41	23		1
<i>Lasioglossum politum</i> (Schenck 1853)	female	Laspolf	1.4						2	1	13	1	15		
<i>Lasioglossum villosulum</i> (Kirby 1802)	female	-	1.9		1					1		1	1		
<i>Lasioglossum zonulum</i> (Schränk 1781)	female	Laszonf	2.8		5		3						8		
<i>Megachile circumcincta</i> (Kirby 1802)	female	-	4.9		1								1		
<i>Megachile willughbiella</i> (Kirby 1802)	female	-	3.7		1								1		
<i>Osmia bicornis</i> (Linnaeus 1758)	female	-	4.0		1		1						2		
	male	Osmbicm	3.5				3		7		8		18		1
<i>Osmia brevicornis</i> (Fabricius 1798)	male	-						1				1			
<i>Osmia caerulea</i> (Linnaeus 1758)	male	-	2.8							1		1			
<i>Osmia cornuta</i> (Latreille 1805)	female	Osmcorf	5.1						2		9		11	1	4
	male	Osmcorm	4.2				6		13	1	47	1	66	2	19
<i>Sphecodes ephippius</i> (Linnaeus 1767)	female	-	1.8							1		1			

Table S4. Bee species collected in pan traps covered with hail net and in open traps. Coloured pan traps were placed in an established row of high stem apple trees in Zurich, Switzerland, during the flowering period 16 March – 8 May 2020. The pots of three trapping stations were covered with hail netting while the pots of one station was open.

Species	Sex	Thorax width [mm]	Open 1 trap	With net 3 traps
<i>Andrena cineraria</i> (Linnaeus 1758)	female	4.1	2	
	male	3.2	2	
<i>Andrena vaga</i> Panzer 1799	female	4.1	16	
	male	3.1	30	
<i>Apis mellifera</i> Linnaeus 1758	worker	4.2	3	
<i>Bombus sylvarum</i> (Linnaeus 1761)	female	6.8	1	
<i>Colletes cunicularius</i> (Linnaeus 1761)	female	4.9	12	
	male	4.1	77	
<i>Halictus scabiosae</i> (Rossi 1790)	female	3.5	7	
<i>Halictus simplex</i> -group Blüthgen 1923	female	2.8	4	
<i>Lasioglossum calceatum</i> (Scopoli 1763)	female	2.8	2	1
<i>Lasioglossum laticeps</i> (Schenck 1868)	female	2.1	1	6
<i>Lasioglossum malachurum</i> (Kirby 1802)	female	2.4	9	10
<i>Lasioglossum pallens</i> (Brullé 1832)	female	2.1	1	
<i>Lasioglossum politum</i> (Schenck 1853)	female	1.6	2	1
<i>Osmia bicornis</i> (Linnaeus 1758)	male	3.1	1	1

Table S5. Hand-collected bees from inside the netted plot. Bees were collected inside the netted plot of apple trees in Zurich, Switzerland, during the flowering period of the ‘Ladina’ and ‘Nicogreen’ trees included in the experiment in the years 2016-2020. Large bees that were seen inside the netted plot when other work in the orchard was done, or after the flower observations, were caught sporadically. In the year 2020 (same setup of the netted plot as described for the years 2018 and 2019), an additional planned hand collection was conducted to collect bees recognized in relatively high numbers at the bottom of the insect side netting inside the netted plot.

Species	Sex	Date	Number
Sporadic hand collections 2016-2019			
<i>Apis mellifera</i> Linnaeus 1758	worker	30.05.2016	1
<i>Apis mellifera</i> Linnaeus 1758	worker	23.06.2016	1
<i>Bombus humilis</i> Illiger 1806	worker	08.05.2018	2
<i>Bombus sylvestris</i> Lepeletier 1832	female	08.05.2018	1
<i>Halictus tumulorum</i> (Linnaeus 1758)	female	27.05.2016	1
<i>Lasioglossum laticeps</i> (Schenck 1868)	female	25.04.2019	1
<i>Osmia bicornis</i> (Linnaeus 1758)	female	24.04.2019	1
Planned hand collection 2020			
<i>Halictus tumulorum</i> (Linnaeus 1758)	female	24.04.2020	5
<i>Lasioglossum glabriusculum</i> (Morawitz 1872)	female	24.04.2020	4
<i>Lasioglossum pauxillum</i> (Schenck 1853)	female	24.04.2020	37
<i>Lasioglossum politum</i> (Schenck 1853)	female	24.04.2020	4
<i>Lasioglossum villosulum</i> (Kirby 1802)	female	24.04.2020	2
<i>Sphecodes puncticeps</i> Thomson 187	female	24.04.2020	1



Figure S1. Aerial photograph of the experimental area. The red rectangle marks the netted plot of apple trees with the arthropod trapping stations 5-8 (green circles), placed in row 6, 5, 4, and 2. The position of the trapping stations 9-12 within the groups of 10 apple trees ('Nicogreen'; A-D) in each direction of the netted plot, are indicated as blue circles together with the distance to the nearest 'Ladina' trees. Inset picture: Trapping station with yellow, blue, and white plastic pot filled with water + detergent to collect arthropods.

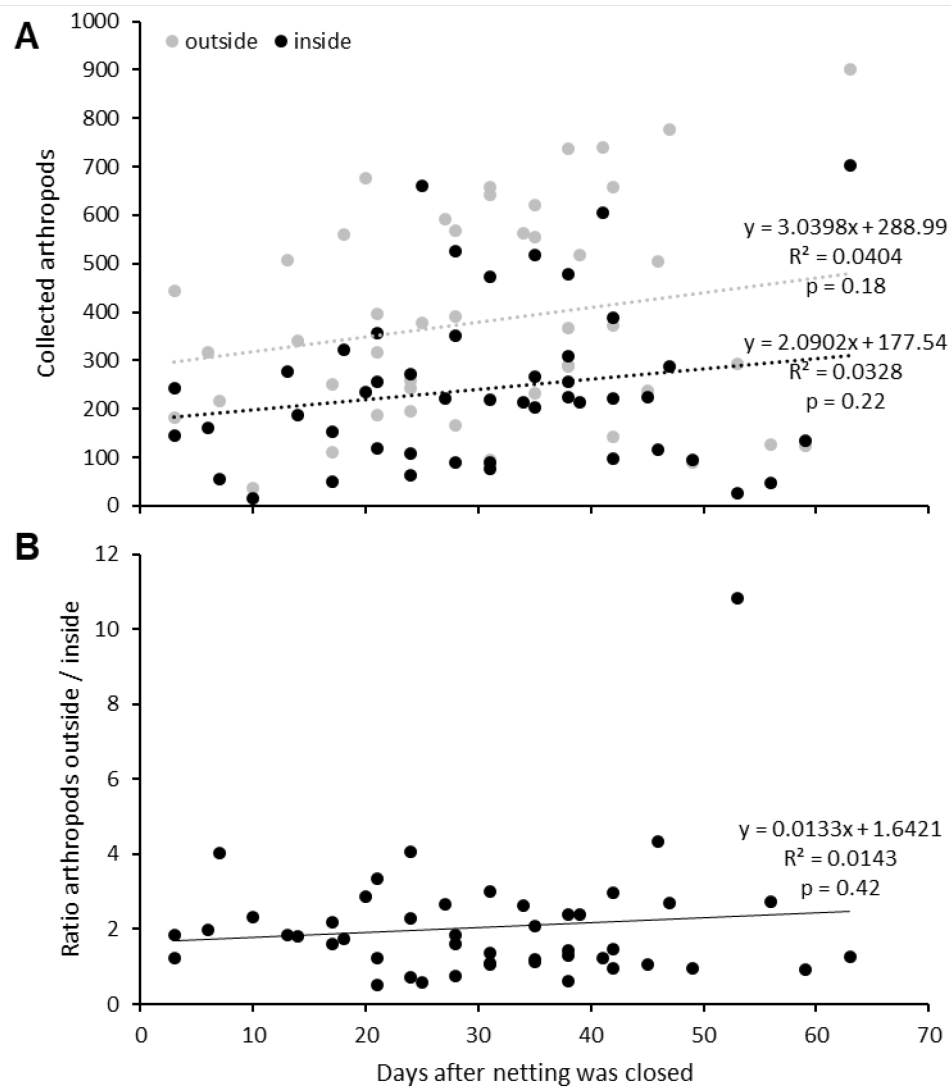


Figure S2. Arthropod numbers per sampling date. A) Arthropods collected outside and inside the netted plot over the years 2016-2019 for individual sampling dates, presented as days after the hail net was closed. B) Ratio of arthropods collected outside / inside. The trend lines represent linear regressions.

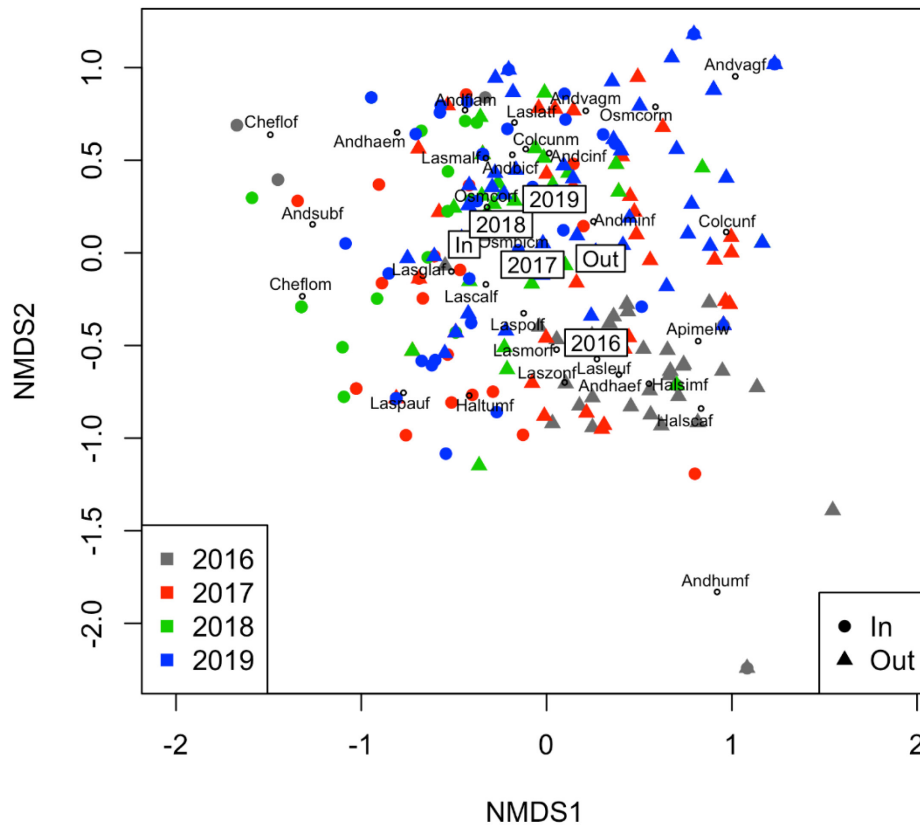


Figure S3. Ordination plot with data from each collection date. Multivariate ordination with non-metric multidimensional scaling (NMDS) of bees collected in coloured pan traps placed inside (circles) and outside (triangles) the netted plot of apple trees during the flowering period of the ‘Ladina’ and ‘Nicogreen’ trees included in the experiment in the years 2016-2019 (different colours). Each data point represents one collection date and one trapping station. Species with less than 5 collected individuals over all years and traps were not considered. Species names are abbreviated with the first 3 letters of the genus and the species name followed by the sex (m = male, f = female, w = worker). The centroids of each year and the plot type (in/out) are indicated in boxes.

A**B**

Figure S4. Net construction and bee nest. A) Net construction at the short sides of the netted plot of apple trees. The hail netting covering the plot, closed by zippers (2018, 2019), was spanned over the beam of the side netting construction. B) Fresh bee nests in the soil near the side netting inside the netted plot.