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RESEARCH ARTICLE – DOSSIER OPEN ACCESS

SUNFLOWER: SOME EXAMPLES OF CURRENT RESEARCH
TOURNESOL : EXEMPLES DE TRAVAUX DE RECHERCHE

DOSSIER

Differential attractiveness of sunflower cultivars to the honeybee *Apis mellifera* L.

Nicolas Cerrutti and Célia Pontet*

Terres Inovia, 1 avenue Lucien Brétignière, 78850 Thiverval Grignon, France

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Abstract – In France, sunflower honey production has decreased since the nineties and professional beekeepers report important honey yield variability between years and locations. Consequently, melliferous potential of current sunflower cultivars is questioned as it is not part of traits held in breeding programs. To investigate this question, a 3-years study was conducted on two sites and attractiveness for honeybees of 13 current sunflower cultivars was assessed under natural conditions. The number of honeybee visits per plant was recorded daily during blooming period on cultivars randomly distributed on experimental plots. Sunflower genetic was a major factor influencing honeybee attendance on plots and discrepancy between most and least visited cultivars reached a factor of 3. Cultivars relative attractiveness was consistent between years and sites. Potential link between sunflower attractiveness for honeybees and its melliferous characteristics is discussed. If confirmed, farmers' choice concerning cultivars at a territory scale could contribute to enhance nectar resource for pollinators and to increase viability of apicultural activity.

Keywords: Honeybee / attractiveness / sunflower / cultivar / nectar

Résumé – Étude de l'attractivité de différents cultivars de tournesol pour l'abeille domestique *Apis mellifera* L. En France, la production de miel de tournesol est en diminution depuis les années 1990 et les apiculteurs professionnels rapportent une importante variabilité de rendements en miel entre les années et les sites. De ce fait, ils s'interrogent sur le potentiel mellifère des cultivars de tournesol actuels comme la production de nectar n'est pas prise en compte dans les programmes de sélection. Afin d'étudier cette question, une expérimentation de 3 ans a été conduite sur deux sites et l'attractivité pour les abeilles de 13 cultivars de tournesol évaluée en conditions naturelles. Le nombre de visites d'abeilles par plante a été enregistré quotidiennement pendant la période de floraison sur les variétés distribuées aléatoirement en placettes expérimentales. La génétique du tournesol s'est révélée être un facteur majeur expliquant le niveau de fréquentation des placettes et les écarts de fréquentation observés entre cultivars ont atteint un facteur de 3. Le niveau d'attractivité mesuré a été relativement constant entre les années et les sites expérimentaux. Le lien entre attractivité pour les abeilles et potentiel mellifère du tournesol est ici discuté. S'il se confirmait, les choix variétaux effectués à l'échelle du territoire par les agriculteurs pourraient contribuer à renforcer la ressource nectarifère disponible pour les pollinisateurs et à augmenter la viabilité de l'apiculture.

Mots clés : Abeille mellifère / attractivité / tournesol / cultivar / nectar

In the eighties, sunflower cultivation in France had a tremendous increase. The arable land devoted to this crop was multiplied by 25, increasing from 38 633 to 1 047 649 ha between 1978 and 1987 (Agreste, 1943–2013). Along with oilseed rape, sunflower represents a major entomophilous crop in France which went along with the development of industrial beekeeping. At present time, a lot of professional beekeepers, especially in the regions Centre and Midi-Pyrénées report that sunflower honey production is irregular between years and locations and below the level they knew in the nineties. Unfortunately, no official data are available to provide an overview of the phenomenon and to confirm what is observed in the field.

A main hypothesis is that poor honey harvests in certain areas could be linked to the cultivation of cultivars with small melliferous potential. Melliferous potential in sunflower cultivars is well studied in Romania in order to help beekeepers to choose their locations and to predict honey yield they can expect according to sunflower cultivar and climatic conditions of the year (Ion *et al.*, 2007). But in France, no references are available for current cultivars according to pedologic and

* Correspondence: n.cerrutti@terresinovia.fr

Table 1. Site location and data collection periods.

Year	2011	2012	2013	
Site	A : Le Magneraud	B : En Crambade	A : Le Magneraud	B : En Crambade
Location	46°07′09.34″ N, 0°50′06.57″O	43°25′25.27″ N 1°39′02′34″ E	46°07′09.70″ N 0°49′49.15″ O	43°25′35.66″ N 1°39′25.18″ E
Soil caracteristics	Shallow Argilo-calcareous	Deep Argilo-calcareous	Deep Argilo-calcareous	Deep Clay-loamy
Data collection periods	June 24th to July 5th	July 3rd to July 19th	July 28th to August 21st	July 16th to august 8th

climatic local context. As melliferous characteristics are not considered in breeding programs, great variability can be expected. Breeding programs particularly aim at improving yield level and stability, developing high autogamous hybrids. We can expect as a result non intentional selection towards less melliferous genotypes. Hence, the management of nectar and pollen resources at a territory scale is of particular interest for pollinators which populations and diversity are severely declining worldwide (Biesmeijer *et al.*, 2006; Goulson *et al.*, 2008; Kluser and Peduzzi, 2007; Potts *et al.*, 2010). Pollen diversity is essential to pollinators' survival and fitness (Di Pascuale *et al.*, 2013; Schmidt *et al.*, 1987). When it comes to nectar, creating cropping systems favorable to pollinators require to diversify rotations, introducing various mass flowering species with long flowering periods such as sunflower, faba bean or buckwheat in order to avoid food shortage which can occur in certain areas and especially before winter (Biesmeijer *et al.*, 2006; Decourtye *et al.*, 2013; Requier *et al.*, 2015). In this context, sunflower is a key to create bee friendly environments. It is a crop bringing nectar and pollen in summertime which allows honeybee colonies to refill their stocks before wintering.

Melliferous potential in a cultivar refers to its ability to produce nectar with high glucidic index which brings a high honey yield (Ion *et al.*, 2007). Nectar accessibility to bees, depending on morphological aspects of the florets such as shape and length must be considered as well. Indeed, flower morphology can represent a physical barrier susceptible to limit pollinator access to nectar resource (Torres and Galetto, 2002). Finally, to be melliferous, a cultivar has to be attractive for honeybees. Visual and olfactive aspects can also play a role in attractiveness in link with nectar reward (Wright and Schiestl, 2009).

On other flower species, literature shows evidence that heritable traits are responsible for variation in plant attractiveness for pollinators (Mitchell, 2003; Mitchell and Shaw, 1993; Wolf *et al.*, 1999). Some physiological traits are directly linked to the nectar resource for example the quantity of nectar produced per floret, the glucidic index, and also the sugar composition (Ion *et al.*, 2007). Other traits are linked to nectar resource accessibility such as corolla length or flower morphology (Atlagić *et al.*, 2003; Torres and Galetto, 2002).

Despite these elements, no studies have been done to assess melliferous potential of sunflower cultivars currently present in France and its variability according to pedoclimatic context. Here is presented a first step to investigate this potential using an original method based on the study of honeybee attendance during flowering period. Attendance, or the number of bee visits on the crop can be considered as an integrative parameter because social bees foraging preferences take in account nectar secretion and pollen quality and resource accessibility (Atlagić *et al.*, 2003; Cook *et al.*, 2003; Cnaani *et al.*, 2006; Tepedino and Parker, 1982).

1 Materials and methods

The study was conducted in one site, Le Magneraud (named A, Central west of France) in 2011 and En Crambade (named B, South West near Toulouse) in 2012, and in both sites in 2013 (Tab. 1). A preliminary study was performed the first year, followed by an in-depth study in 2012 and 2013.

1.1 Preliminary study

Forty-five sunflower cultivars were sown randomly in microplots with 3 replicates. Each plot was 32 m^2: 10 m long with a row space of 53 cm. Four *Apis mellifera* L. colonies/ha were settled on the field margin at a distance of 10 m from the first rows. Then, during 11 successive days, honeybee visits were recorded twice a day (10 A.M., 15 P.M.) by counting the number of bees foraging on each microplot during 1 min.

1.2 In-depth study

Further studies were conducted in 2012 (site B), and in 2013 (both sites). On each trial, 13 cultivars were sown. Eleven of them were chosen according to results of preliminary study. Two new cultivars (2 and 8) were added in this study. On each trial, the 13 cultivars were sown randomly with 3 replicates (1248 m^2). The surroundings of the trial were planted with a mix of the 13 varieties on a surface of 1300 m^2 to avoid blooming period discrepancy between the measured plots and the surroundings. This precaution was taken in order to maximize honeybee attendance on experimental plots and to facilitate observations. On each experimental plot, the number of plants was equalized to 5.5–6 plant per meter. Then, the number of plants was recorded on each plot. Attractive sunflower heads and foraging honeybees were daily counted manually on each plot from the beginning to the end of flowering period. A capitula was considered as attractive when ligulated flowers started to open until all the florets were wilted. To increase foraging density, four *Apis mellifera* L. colonies/ha were settled on the field margin one week before flowering occured.

Table 2. Earliness at flowering for each cultivar according to the trials.

Earliness classes		1	2	3
2012 site B	Dates	July 2nd and 3rd	July 4th, 5th and 6th	July 7th and 8th
	Cultivars	3, 5	2, 4, 7, 9, 10, 11, 12, 13	1, 6, 8
2013 site B	Dates	July 18th to 23rd	July 24th to 27th	
	Cultivars	3, 5, 12, 13	1, 2, 4, 6, 7, 8, 9, 10, 11	
2013 site A	Dates	July 31th to August 2nd	August 3rd to 4th	
	Cultivars	2, 3, 5, 9, 12	1, 4, 6, 7, 8, 10, 11, 13	

1.3 Methodological aspects

For data analyses, statistics were performed on cumulative attendance values of cultivars rather than on daily counts which were found to be non-relevant for comparisons because of: (1) earliness discrepancy between cultivars; (2) variations in climatic conditions and (3) their interactions which contributes to create inequivalent condition for assessment.

1.4 Attendance Index calculation

To summarize honeybee attendance on each cultivar during blooming period, an attendance index was calculated. Expressed in percentage, it represents the total number of honeybee visits in the cultivar during the blooming period relative to the mean number of visits in the whole trial (one site in a given year). This index is calculated as follows:

$$\text{Attendance index}_{\text{Var}A} = \frac{N \times S\ (A)}{S\ (A) + S\ (B) + \ldots + S\ (N)} \times 100$$

N = number of cultivars

$S\ (A)$ = Mean cumulative number of bees/plant for variety A during blooming period.

1.5 Earliness at Flowering

The flowering initiation dates: opening of ligulate flowers were recorded for cultivars in each trial. Then cultivars were classified in different groups (1, 2 or 3) according to these dates. Class 1 refers to the earliest flowering cultivars. Classification is shown in Table 2.

1.6 Statistical analyses

Statistical analyses were performed using SAS 9.4. The variable of interest is the mean cumulative number of bees.

The linear model is described as follows.

$$X = \text{Cultivar} + \text{year\_site} + \text{Replicate(year\_site)} + \text{Earlinessclass}.$$

Spearman's correlation tests were performed to assess the consistency of attractiveness results according to the year and site.

Eventually, the influence of climatic conditions (temperature and rainfall) on daily honeybee attendance on plots was assessed using another linear model:

$$Y = \text{temp} + \text{Precipitation}.$$

2 Results

2.1 Flowering durations

Flowering durations were, on average, 22 days in 2013 for site A, 17 and 24 days in 2012 and 2013 respectively for site B (Fig. 1). Values ranged from 11 to 21 days respectively in site B 2012 and site A 2013. Flowering duration appeared to be under the influence of the year (one way ANOVA, $p < 0.0001$), and the site ($p = 0.0136$). However, the genetics had no significant influence on this variable ($p = 0.99$).

2.2 Honeybee visits and flowering dynamics

Honeybee attendance is highly correlated to the percentage of attractive sunflower heads in experimental plots (Fig. 2, spearman correlation test, $r^2 = 0.98$). It shows that the number of bees counted is closely linked to the blooming stage of the crop. At the scale of a trial and when expressed in function of the date, these variables design a bell curve showing that honeybee attendance follows temporal variation of the resource available in the field (Fig. 3).

2.3 Honeybee visits and sunflower genetics

In the preliminary study, Attendance Index ranged from 30 to 218% of the trial mean value (Fig. 4). Cultivar number 34 was 7 times more visited that cultivar number 5. Mean Standard deviation between blocs was 4.16%, which is relatively low, indicating that attendance results between replicates is very consistent. The cultivars, whenever their spatial position in the field present very close values of attendance.

In in-depth studies, attendance index values ranged from 47 to 148% (Fig. 5). During these experiments, the gap between extreme values was less important than in the preliminary study. Statistical analyses revealed that attendance on plots was significantly influenced by year_site ($p < 0.0001$), the replicate ($p < 0.0134$) and the cultivar ($p < 0.0001$). On the opposite, cultivar earliness had no influence on attendance ($p = 0.69$). It confirms the significant influence of sunflower genetics on the level of attractiveness for honeybees and allows excluding the hypothesis of a confounding effect with earliness.

Beyond factors inherent to the plants, temperatures and precipitations have an impact on daily attendance on the plots ($p = 0.0172$ and $p < 0.0001$ respectively).

Cultivars visitation by honeybees for each trial is summarized in Table 3 and Figure 6. Consistency of results between

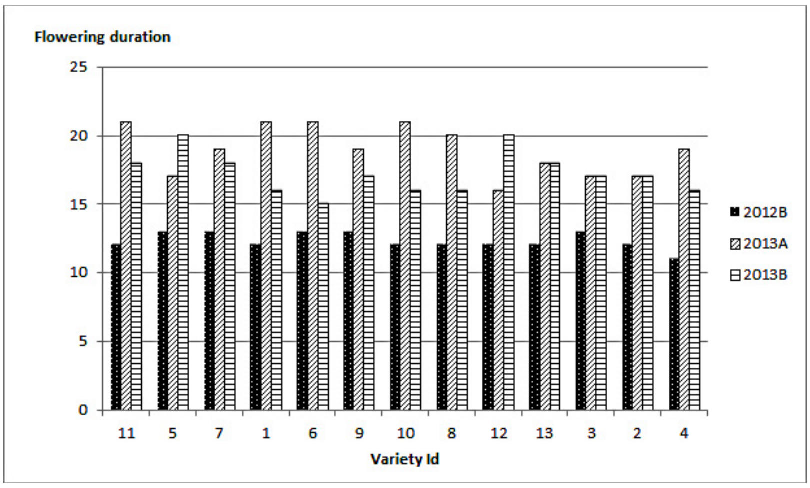

Fig. 1. Flowering durations of cultivars according to the year and site.

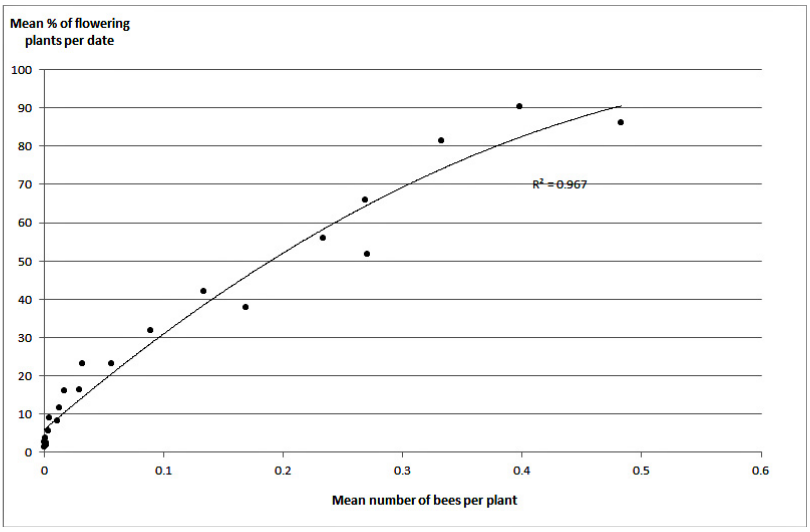

Fig. 2. Honeybee attendance in function of the number of plants with open ligulated flowers (trial= 2013 site A) – all varieties are represented.

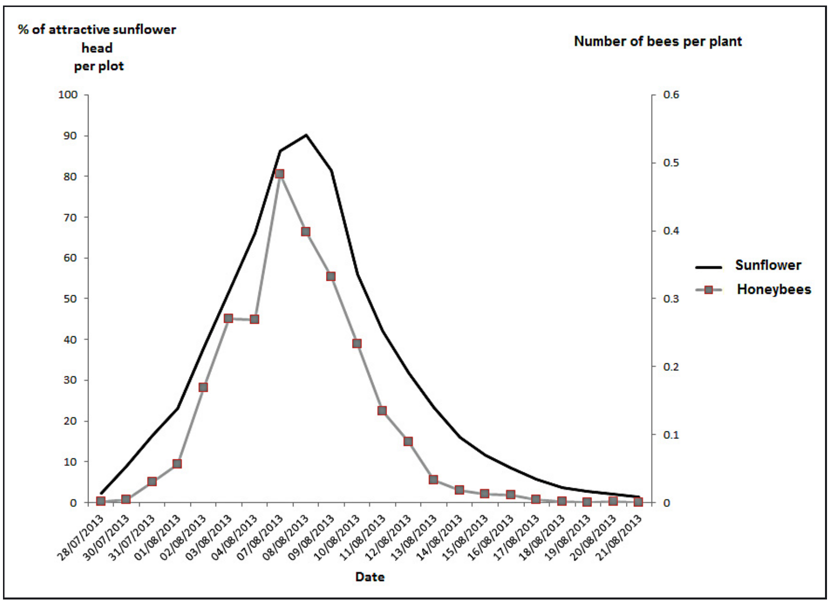

Fig. 3. Number of bees per plant and % of attractive sunflower heads according to the date (trial = 2013 site A).

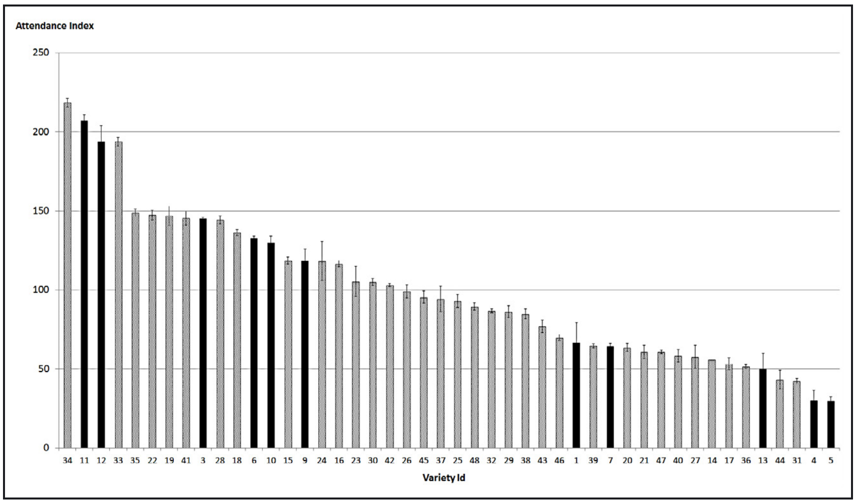

Fig. 4. Attendance index values for cultivars assessed in preliminary study. Black colored bars correspond to cultivars assessed in 2012 and 2013.

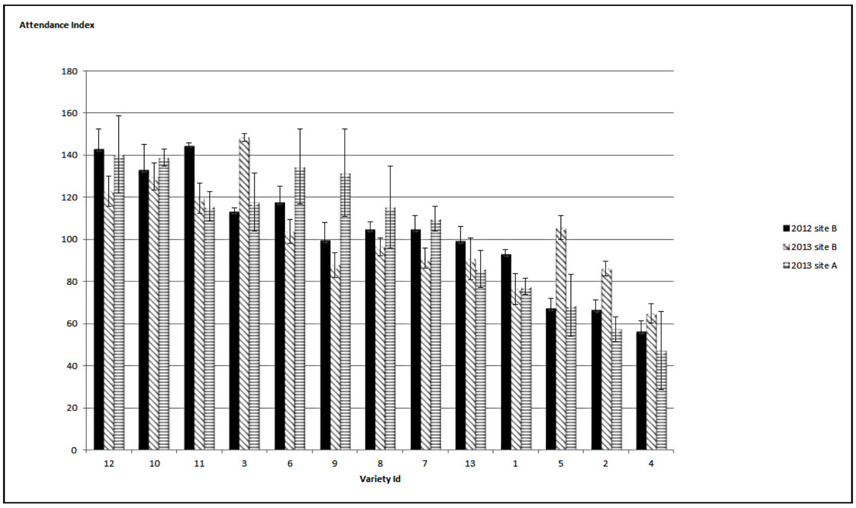

Fig. 5. Attendance index values for cultivars assessed in 2012 and 2013 trials. Cultivars 2 and 8 were not assessed in the preliminary study.

Table 3. Mean cumulative number of bees per plant during the blooming period according to year and site.

Variety Id	2013 site B	2013 site A	2012 site B
12	1.74	3.47	3.52
10	1.84	3.43	3.29
6	1.47	3.33	2.90
9	1.24	3.26	2.46
3	2.10	2.91	2.80
11	1.69	2.86	3.57
8	1.37	2.85	2.58
7	1.29	2.71	2.59
13	1.28	2.13	2.45
1	1.08	1.92	2.30
5	1.49	1.70	1.66
2	1.22	1.42	1.64
4	0.92	1.16	1.39

Table 4. Results of spearman correlation tests performed on attendance data from Table 3.

	2013 site B	2013 site A	2012 site B
2013 site B	1	0.7 $p = 0.008$	0.78 $p = 0.0017$
2013 site A	0.7	1	0.87
2012 site B	0.78	0.87 $p = 0.0001$	1

trials is very high for cultivars 2, 4 and 12 but is very low for other cultivars, especially the cultivar number 3. Spearman's correlation tests shown in Table 4 demonstrate that the relative rating between trials is consistent even if some cultivars seems to perform differently probably because of genetic x environment interactions.

3 Discussion

Sunflower cultivars assessed in this study are differently attractive for honeybees and earliness at flowering is not

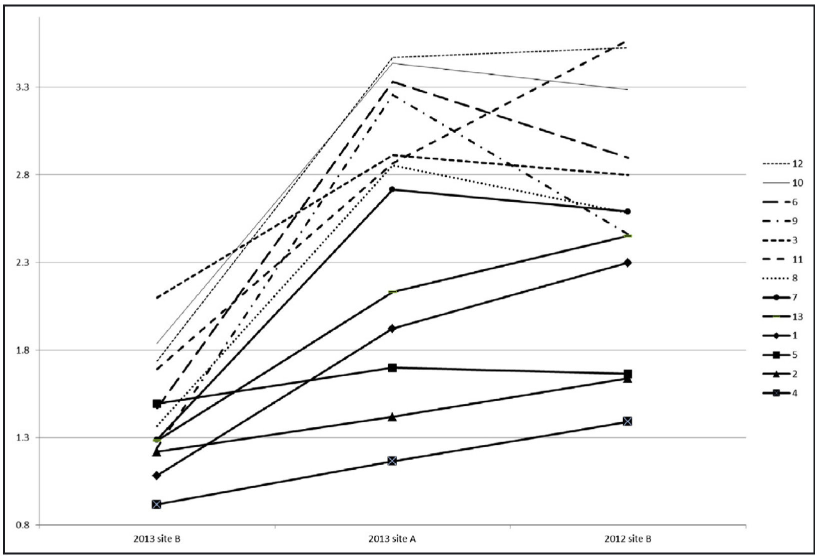

Fig. 6. Mean cumulative number of bees per plant registered for each cultivar according to year and site.

responsible for the results obtained. Consistency of attendance results between years and locations suggests that Honeybees show stable preference for certain cultivars even if genotypes x environment interactions are to be expected. Although, it is remarkable that noneof the tested cultivars was totally neglected by the insects. However, this selective foraging behavior raises the question of variation in melliferous potential among genotypes *i.e*. the quality and quantity of nectar provided to honeybees and its accessibility. In the following discussion we will focus on nectar resource because it is involved in honey production and has economic implications even if pollen quality may also influence sunflower attractiveness (Cook *et al.*, 2003).

Frequentation of sunflower plots is the result of colonies' foraging activity which is based on several behavioural aspects at individual scale and in interaction with peers: (1) visual detection of sunflowers by foragers which is a combined perception of colors, morphological aspects of the capitulates and floral sent; (2) perception of the nectar reward; (3) communication of the food source to other foragers *via* waggle dance whose characteristics influences the intensity of recruitment. It has been shown that selection of nectar sources by colonies is not the consequence of a centralized collective decision but is simply the result of an increase in foragers recruitment on most profitable sources via modification of waggle dance (De Marco and Farina, 2001; Seely *et al.*, 2000). In this study, plots are so closed from each other's and the design is so intricate that it seems difficult to consider that attendance levels are the result of recruitment variation inside the field; but it appears clearly that honeybees have the ability to detect and forage preferentially on certain cultivars to the detriment on the others which suggests that they provide more profitable food sources.

For honeybees, nectar profitability is function of nectar concentrations in sugar, sugar composition, but also time spent to collect it which depends on its accessibility in flowers (Wainselboim *et al.*, 2002). Accessibility requires suitable morphological aspects for florets including corolla length, position and shape of stamina etc. As these traits have no agronomic interest, and so many criterions have to be taken into account for pathogen resistance, yield, oil content. . . , a great variability can be expected concerning profitability of nectar sources in current cultivars. Moreover, with the concern to increase yield stability in the majority of pedoclimatic conditions, sunflower breeding programs may have led to a reduction of their dependence towards entomophilous pollination for grain production. Indeed, high insect dependence for pollination in sunflower represents no value for farmers because it increases crop vulnerability to bad weather condition during blooming period. As a result we could even make the hypothesis that sunflower traits favorable to pollinators have been indirectly counter-selectionned.

Further studies would be necessary to determine sunflower traits which are responsible for attractiveness and their prevalence. But, if linked to melliferous potential, as some studies suggests (Rabinowitch, 1993; Shykoff *et al.*, 1995; Southweek *et al.*, 1981), sunflower attractiveness would be a major lever to activate in order to increase the capacity of a territory to host pollinators and especially *Apis mellifera* L. Indeed, in certain areas where semi-natural habitats are little represented, mass flowering crops such as sunflower are of particular interest. As a result, maximizing nectar resource at a territory scale by cultivating melliferous sunflower cultivars would have positive implications by increasing honey production and viability of apicultural economic activity and as well as food sources available for all pollinators in the global context of their decline.

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References

Agreste, 1943–2013. Situation mensuelle départementale grandes cultures : tournesol.

Atlagic J, Joksimovic J, Sakac Z, Miklic V, Dusanic N. 2003. Mode of inheritance and heritability of disc flower corolla length and nectar content in sunflower. *Genetika* 35: 59–65.

Biesmeijer JC, Roberts SPM, Reemer M, *et al.* 2006. Parallel declines in pollinators and insect-pollinated plants in Britain and the Netherlands. *Science* 313: 351–354.

Cnaani J, Thomson JD, Papaj DR. 2006. Flower Choice and Learning in Foraging Bumblebees: Effects of Variation in Nectar Volume and Concentration. *Ethology* 112: 278–285.

Cook SM, Awmack CS, Murray DA, Williams IH. 2003. Are honey bees' foraging preferences affected by pollen amino acid composition? *Ecol. Ent.* 28: 622–627.

De Marco R, Farina WM. 2001. Changes in food source profitability affect the trophallactic behavior of forager honeybees (*Apis mellifera*). *Behav. Ecol. Sociobiol.* 50: 441–449.

Decourtye A, Gayrard M, Chabert A, *et al.* 2013b. Projet Polinov, épisode 2 – Vers des systèmes de cultures favorables aux abeilles. *Phytoma* 667: 11–14.

Di Pasquale G, Salignon M, Le Conte Y, *et al.* 2013. Influence of Pollen Nutrition on Honey Bee Health: Do Pollen Quality and Diversity Matter? *PLoS One* 8: 8.

Goulson D, Lye GC, Darvill B. 2008. Decline and conservation of bumble bees. *Annu. Rev. Ent.* 53: 191–208.

Ion N, Stefan V, Ion V, Fota G, Coman R. 2007. Results concerning the melliferous characteristics of the sunflower hybrids cultivated in Romania. *Animal Sci. Biotechnol.* 40: 2.

Kluser S, Peduzzi P, 2007. Global Pollinator Decline: A Literature Review. [Mandate from:] United Nations Environment Programme. United Nations Environment Programme.

Mitchell RJ. 2004. Heritability of nectar traits: why do we know so little? *Ecology* 85: 1527–1533.

Mitchell RJ, Shaw RG. 1993. Heritability of floral traits for the perennial wild flower *Penstemon centranthifolius* (Scrophulariaceae): clones and crosses. *Heredity* 71: 185–192.

Potts SG, Biesmeijer JC, Kremen C, Neumann P, Schweiger O, Kunin WE. 2010. Global pollinator declines: trends, impacts and drivers. *Trends Ecol. Evol.* 25: 6.

Rabinowitch HD, Fahn A, Meir TAL, Lensky Y. 1993. Flower and nectar attributes of pepper (Capsicum annuum L.) plants in relation to their attractiveness to honeybees (*Apis mellifera* L.). *Annu. Appl. Biol.* 123: 221–232.

Requier F, Odoux J-F, Tamic T, *et al.* 2015. Honey bee diet in intensive farmland habitats reveals an unexpectedly high flower richness and a major role of weeds. *Ecol. Appl.* 25: 881–890.

Schmidt JO, Thoenes SC, Levin MD. 1987. Survival of Honey Bees, Apis mellifera (Hymenoptera: Apidae), Fed Various Pollen Sources. *Annu. Entomol. Soc. Am.* 80: 176–183.

Seeley T, Mikheyev AS, Pagano GJ. 2000. Dancing bees tune both duration and rate of waggle-run production in relation to nectar-source profitability. *J. Comp. Physiol. A* 186: 813–819.

Shykoff JA, Bucheli E. 1995. Pollinator Visitation Patterns, Floral Rewards and the Probability of Transmission of Microbotryum Violaceum, a Veneral Disease of Plants. *J. Ecol.* 83: 189–198.

Southwick EE, Loper GM, Sadwick SE. 1981. Nectar Production, Composition, Energetics and Pollinator Attractiveness in Spring Flowers of Western New York. *Am. J. Bot.* 68: 994–1002.

Tepedino VJ, Parker FD. 1982. Interspecific differences in the relative importance of pollen and nectar to bee species foraging on sunflowers. *Environ. Ent.* 11: 246–250.

Torres C, Galetto L. 2002. Are nectar sugar composition and corolla tube length related to the diversity of insects that visit Asteraceae flowers? *Plant Biol.* 4: 360–366.

Wainselboim AJ, Roces F, Farina WM, 2002. Honeybees assess changes in nectar flow within a single foraging bout. *Animal Behaviour* 63: 1–6.

Wolf S, Lensky Y, Paldi N. 1999. Genetic Variability in Flower Attractiveness to Honeybees (*Apis mellifera* L.) within the Genus Citrullus. *HortScience* 34: 860–863.

Wright GA, Schiestl FP. 2009. The evolution of floral scent: the influence of olfactory learning by insect pollinators on the honest signaling of floral rewards. *Funct. Ecol.* 23: 841–851.

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