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RESEARCH ARTICLE

Animal-pollinated crops and cultivars—A quantitative assessment of pollinator dependence values and evaluation of methodological approaches

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Abstract

1. Crop pollinator dependence (PD) values are key when assessing a pollinator's contribution to agriculture, guiding management plans and policies for sustainable crop production. However, the available global compilations of crops PD are outdated and neglect variability between related crops and accessions (variety/cultivar), as well as pollen limitation (PL), that is the production lost due to inadequate pollen receipt.
2. Here, we obtained quantitative PD values for animal-pollinated crops and their accessions, using data from available pollination experiments worldwide. We also tested pollination methodologies to assess their impact on PD values and to define suitable methodological guidelines for future pollination studies.
3. We provide a list of continuous PD values for 141 crops, including 317 accessions and 37 crops not listed in previous assessments. We found that, globally, 74% of animal-pollinated crops are highly dependent on pollinators, and more than 40% of their production is associated with animal pollination. Pollen limitation was detected in 51% of the dataset entries, indicating that estimates calculated with open pollination studies underestimate crop pollinator dependence and, therefore, do not represent the true contribution of pollinators to food production.
4. *Synthesis and applications*: Commonly applied methods for assessing PD values can lead to underestimations. Future studies evaluating pollinator dependence levels of crops and their accessions (i.e. potential pollinator contribution) should consider the possibility of pollen limitation in the study site, incorporating hand pollen supplementation (to open flower), open pollination, and pollinator exclusion treatments, preferably using the whole plant or branch as the unit of assessment. The PD values provided here, from studies that allow the incorporation of the concept of pollen limitation, enable more accurate quantifications of pollinator contribution to crop production. These PD values are an invaluable baseline and a requirement for future accurate evaluations of the

value of pollinators for food security, supporting pollinator-friendly practices in agroecosystems.

KEYWORDS
agriculture, animal pollinator contribution, crop yield, ecosystem service, hand pollen supplementation, pollen limitation, pollination

1 | INTRODUCTION

Biotic pollination is a crucial biodiversity-dependent ecosystem service that contributes to crop yield, supports food security and provides other ecosystem services (Dicks et al., 2021; Power, 2010). Together with managed pollinators, diverse and abundant wild pollinator communities ensure the reproduction of pollinator-dependent crops, increasing yields and/or improving the quality of fruit and seeds, even in self-compatible crops (Klatt et al., 2014; Klein et al., 2003). Unfortunately, there is evidence of pollinator decline, driven primarily by human-induced changes, and pollination services may be at risk, with implications for food security and human well-being (Dicks et al., 2021; Potts, Imperatriz-Fonseca, et al., 2016).

The ability of a given crop field to achieve its maximum production potential depends on numerous environmental factors, such as availability of nutrients and water, biotic interactions, and pest levels (Licker et al., 2010). For pollinator-dependent plant crops that have as their primary product fruits or seeds, pollination is directly linked with crop yield. In these crops, yield is mainly the result of two components (Figure 1): (1) crop auto-pollination ability (the ability to produce fruits and/or seeds in the absence of pollination vectors, Figure 1—AUTO bar) and (2) pollination services available at each place and time (open pollination, Figure 1—OPEN bar). Altogether, they result in yields that, under optimal conditions, are theoretically equal to (3) the production under optimal levels of pollination (Figure 1—OPT bar).

The difference between open and optimal yields is known as pollen limitation (PL; Figure 1) and is caused by insufficient and/or inefficient pollination services (Bartomeus et al., 2014; Toledo-Hernández et al., 2017). Following Liebig's law (Von Liebig, 1840), crop yield is determined by the most limiting factor. In pollinator-dependent crops, when no other factors limit yield (as expected in optimized agricultural systems), pollination service may be the limiting factor (Tamburini et al., 2019). PL leads to reduced productivity through a quantitative reduction in the amount of a crop produced and/or a loss in crop quality (Vaissière et al., 2011).

The contribution of animal pollinators to crop yields (Figure 1) can vary significantly due to spatial, temporal, and biotic factors (Bishop & Nakagawa, 2021; Mallinger et al., 2021; Webber et al., 2020). Pollinator communities are largely impacted by factors such as regional biodiversity, landscape structure, environmental conditions during flowering and local management practices (Holland et al., 2017; Mota et al., 2022; Potts et al., 2010; Senapathi et al., 2017) and, consequently, pollination services provided by pollinator communities are likely to show significant variation.

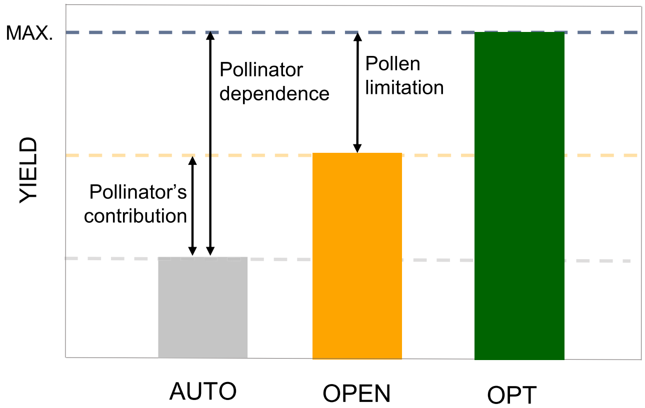

FIGURE 1 Theoretical representation of pollination components associated with yield in pollinator-dependent crops: autonomous self-pollination levels (AUTO), open levels of pollination (OPEN), and optimal pollination levels for local study conditions (OPT). Associated indexes are also presented: (1) pollinator's contribution, yield associated with existing pollination services; (2) pollinator dependence, yield directly dependent on pollinators (for simplification, here we considered a crop with negligible wind pollination contribution) and (3) pollen limitation, yield loss associated with limited pollen deposition levels. See Box 1 for methodologies associated with estimations of each component.

The relative difference in yield resulting from crop auto-pollination ability and optimal pollination corresponds to the potential pollinator's contribution to production, that is the true level of PD, a metric widely used to endorse the importance of pollinators to humans (Figure 1). Estimates of the contribution of pollinators to agricultural production can guide both farm management practices and policy making regarding pollinator conservation (Potts, Ngo, et al., 2016). PD values are tools to guide farmers towards practices that improve pollinator communities, benefiting crop yield. For crops highly reliant on animal pollinators, implementing management strategies tailored to protect, sustain and, if needed, attract pollinators to the crop field becomes essential. These strategies typically prioritize the reduction of agrochemicals usage and the promotion of floral resources, habitat connectivity, and nesting sites (Bartomeus et al., 2014; Mota et al., 2022; Potts et al., 2010). Furthermore, by combining the crops PD with their economic value, we can assess the direct economic impact of pollinators on crop production and markets (Gallai et al., 2009; Potts, Imperatriz-Fonseca, et al., 2016; Silva et al., 2021).

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Studies such as Free (1993) and Klein et al. (2007) widely assessed the pollinator's dependence of crops. Klein et al. (2007), the most comprehensive and widely used study, compiled PD values in four categories ("little", "modest", "high" and "essential") for 91 major global crops. This index constitutes the base for current economic assessments of pollination value at regional, national and global scales, facilitating conservation actions and initiatives focused on pollinators and their importance (e.g. Aizen et al., 2009; Gallai et al., 2009; Millard et al., 2023; Potts, Ngo, et al., 2016). However, due to the continuous emergence of crops and the availability of new studies, a revision on PD levels of crops is currently needed. Recent syntheses after the seminal work of Klein et al. (2007) include PD values for emergent crops; nevertheless, they are usually focused on a few economically important crops or specific regions of the globe (see Bishop & Nakagawa, 2021; Giannini et al., 2015; Mallinger et al., 2021). Additionally, within a crop, different accessions (plants that share similar and/or selected traits, including cultivars, varieties and other infraspecific taxonomic levels) may differ greatly in self-compatibility and auto-pollination ability (e.g. Kendall et al., 2020; Klatt et al., 2014), having, hence, different PD levels (e.g. Bishop & Nakagawa, 2021; Carvalheiro et al., 2010; Marini et al., 2015). However, detailed information about PD levels in crop's accessions is scattered in the literature, making it difficult to compile and to our knowledge, it is seldom accounted for in global studies.

Despite the growing availability of studies quantifying PD, there are challenges with currently used methodologies, which could be underrepresenting pollinator importance and their associated economic value. The crops' PD literature generally evaluates production after open pollination, comparing it with the output after pollinator exclusion (Figure 1). Consequently, PD values using open pollination will vary according to local pollinator communities. Therefore, we propose that hand pollen supplementation is more suitable for estimations of PD, as open pollination may lead to underestimations of PD values. For example, for the same plant species, an estimation of PD based on an open pollination reference in an impoverished landscape with unfavourable conditions for pollinators will generate lower PD values than a similar experiment run in a landscape with rich and abundant pollinator communities able to provide suitable pollination services. Because PL is common in wild plants and crops (Bennett et al., 2018; Olhnuud et al., 2022; Potts, Ngo, et al., 2016; Sáez et al., 2022), we expect lower estimates of PD using open pollination than with hand pollination. Moreover, as flower manipulations may affect flower and fruit development (e.g. Hedhly et al., 2009), we expect different methodologies associated with hand pollen supplementations (e.g. emasculation and/or bagging of flowers) to impact PD estimates negatively. On the contrary, experiments conducted on a smaller scale, such as individual flower level, may overestimate PD levels. This can be attributed to resource allocation, where a successfully pollinated flower, such as in hand pollen supplementations, triggers a reallocation of resources, favouring higher-quality pollinations compared to other flowers of the plant (Wesselingh, 2007). Thus, PD values are expected to be higher when pollination treatments are performed at smaller scales (e.g. flower level) than at larger ones (e.g. plant level).

We gathered information on pollination experiments for animal pollinated crops to test the aforementioned expectations and propose a methodological framework to estimate the crops PD. Finally, we provide a list of continuous PD values for animal pollinated crops, including crop accessions when available. We believe that this list can support more accurate economic assessments of the contribution pollinators make to food production at local, regional, and global scales and guide policymaking and farm management practices regarding pollinator conservation.

2 | MATERIALS AND METHODS

2.1 | Dataset development

To assess the contribution of animal pollination to crops production, we used data focused on pollination experiments performed in agricultural contexts and open conditions, from the PolLimCrop dataset (Siopa et al., 2023). The dataset was based on a list of animal pollinated plant crops from which fruit and/or seeds are used as food and goods (FAO's list of crops, available at [https://www.fao.org/faostat/en/#data/QCL (2021)]).

To build a dataset of crops' PD, we selected entries that included three treatments. First, a hand pollen supplementation treatment, in which the flowers were pollen supplemented to achieve optimal pollination. Second, an open pollination treatment, where flowers received pollination services naturally, from the environment. Third, a pollinator exclusion treatment, in which the flowers were excluded from animal pollination by caging or bagging. From the dataset, we also retrieved other variables such as the species and common names of the crop and part of the crop economically used (fruit or seed). From the selected studies, we extracted production results associated with pollination treatments (fruit set, fruit weight, seed set, seed number and/or seed weight), as well as descriptive variables of the study (species and crop name, family, plant accession, crop part used economically, year and scale of the experiment and supplement type; see Table S1 in Supporting information). The study did not require ethical approval.

2.2 | Pollinator dependence estimation

The PD values were calculated using the following equation:

$$\text{PD} = 1 - \left[\text{pollinator exclusion production} / \text{pollinator-associated production}\right]$$

where pollinator exclusion production refers to the production in the absence of pollinators, and pollinator-associated production refers to the production associated with animal pollinator visitation (i.e. hand pollen supplementation or open pollination).

PD estimates were computed using the production variables derived from the commercially used parts (seed and/or fruit). In fruit crops, fruit-related production variables were used, that is fruit set and fruit weight. For seed crops, the seed set and seed number and weight were used, in addition to fruit set. In some cases, where both are economically used, the production variables related to fruits and seeds were used. When several production variables were provided, a mean value of the obtained PD values was calculated. Two PD values were calculated for each entry, one using hand pollen supplementation and pollinator exclusion treatments (PD-SUP) and the other using open pollination and pollinator exclusion treatments (PD-OPEN). PD ranged from 0 to 1, with 0 representing lack of PD and 1 representing the highest level.

A final PD value was obtained for each entry (defined here as PD-final), using either hand pollen supplementation or open pollination treatment, by selecting the maximum value obtained. Variation in production variables is expected, and therefore, cases where open pollination overcomes hand pollen supplementation may occur. For every database entry, PD-SUP, PD-OPEN and PD-final was added to the dataset for the subsequent statistical analyses.

2.3 | Statistical analysis

A total of 170 studies were selected for the calculation of PD, containing hand pollen supplementation, open pollination and pollinator exclusion, and were included in statistical analyses. To compare PD levels after open pollination and hand pollen supplementation, general linear mixed-effects models (GLMMs) were created using PD values from both treatments, with "treatment type" as an explanatory variable. To account for the variation associated with crop identity, "crop" was included as a random variable in all models. Similarly, the "study code" was also used as a random variable to eliminate confounding effects of within-study aspects.

To evaluate if PD values depended on specific aspects of the methodologies used, analyses were performed using PD-final obtained in our dataset. In particular, GLMMs were performed to analyse the effects of the hand pollen supplementation methodology and the scale of the pollination experiment on PD values. The hand pollen supplementation methodology included four techniques (see Table S3, 'supplement type'). The scale included four experimental scales (see Table S3, 'scale'). Again, "crop" and "study code" were used as random factors. GLMMs were performed using the "lmer" function of the R package "lme4" (Bates et al., 2014), with logit transformation of adjusting factor of 0.01 of the R package "car" (Fox & Weisberg, 2019). Model fitting and validation were performed by comparison of the AIC values of the different models and by graphical analysis of the residuals (Zuur et al., 2009). Wald chi-square analyses were used to calculate the effect of tested variables on PD values. We then ran post hoc pairwise comparisons to test for differences within treatments of supplement type and scale, using R package "emmeans" (Length et al., 2018). The studies on apples constituted 33% of PD values in all performed analyses (see Table S2, Crop "Apple"). To test if such a large set of studies on one crop influenced our results, all analyses without apple's entries were rerun.

To allow comparison with previous global studies, we grouped our continuous PD values into the classes used by Klein et al. (2007; little: 0–0.09 PD, modest: 0.10–0.39, high: 0.40–0.89, essential: 0.90–1.00). To identify duplicates, we compared attributes within the analysed dataset, specifically, crop, year of the experiment and associated production values of the included treatments. All analyses and graphs were obtained in R software (version 4.2.1).

2.4 | Pollinator dependence—Compilation table

To provide a comprehensive list of PD values for animal pollinated crops and their accessions, we created a 'Compilation table' (Table S2) containing the mean PD values calculated for the 170 studies used in statistical analyses, and a set of 65 studies reporting only hand pollen supplementation or open pollination (thus excluded from statistical analyses). A full list of contributing studies is given in the Supporting Information.

Mean values were obtained using PD-final of each available entry, plus PD-final of the additional studies (Table S2). The PD values ranged from 0 to 1, with negative values considered as 0, indicating no animal pollinator dependence. Treatments that contributed to mean PD values (either hand pollen supplementation treatment, open pollination, or both) are indicated in the dataset. Similarly, mean PD values were obtained and assembled for all available accessions within the crops (Table S3).

3 | RESULTS

3.1 | Open pollination versus hand pollen supplementation

A total of 170 studies, covering 91 different crops, were used in statistical analyses, including 563 entries with PD values (representing different crops, accessions, years, and experimental sites). A map with the geographical distribution of these studies and entries is provided (Figure 2a). Study type distribution (e.g. article, thesis or proceeding) is provided in the Supporting Information (Table S4). Crops with the most entry values of PD were apple, oilseed rape and almond (representing 33.2%, 6.2% and 4.6% of total entries, respectively). Twenty-seven crops were represented by one value of PD only.

PD values estimated after hand pollen supplementation-associated production were significantly higher (ca. 4.7% higher on average) than those estimated after open pollination ($x^2 = 7.375$, $p = 0.007$; Figure 2b; Table S4). Hand pollen supplementation gave higher PD values than open pollination in 50.8% of cases (Figure 2c, Figure S1a). Hand pollen supplementation and open pollination gave similar PD values in 23.4% of cases (Figure 2c, Figure S1a). Finally, hand pollen supplementation led to lower PD values than open pollination in 25.8% of cases (Figure 2c, Figure S1a).

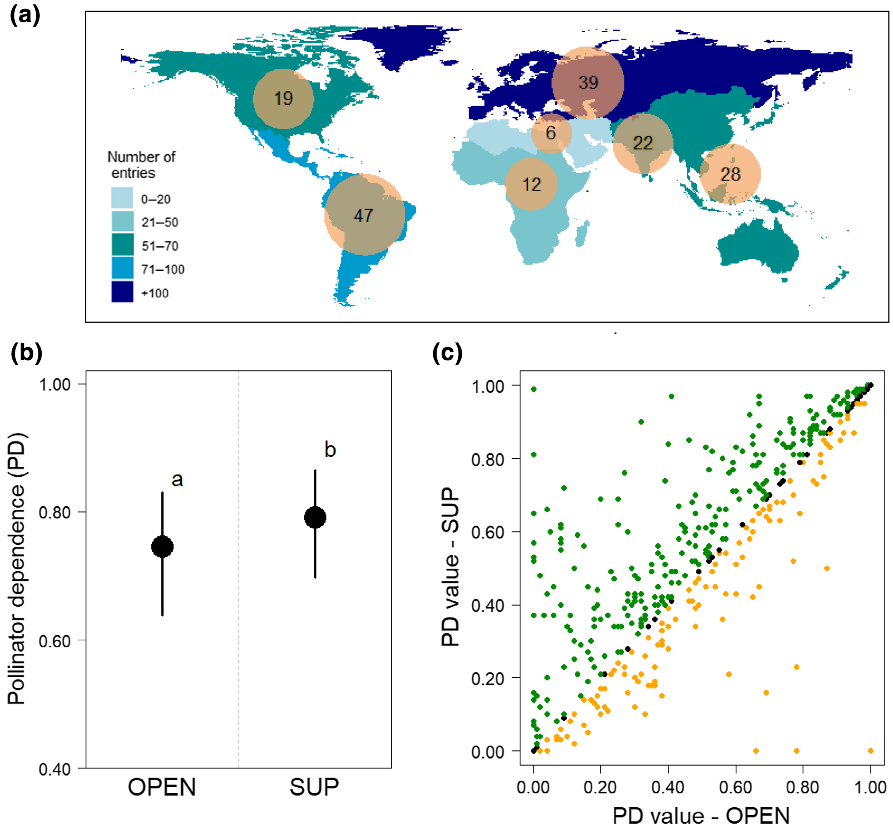

FIGURE 2 (a) Global distribution of data entries and studies of the analysed dataset. The map's colour gradient represents the total number of entries across regions, by ranges. Orange circles represent the total number of studies across regions. (b) Estimated means and 95% confidence intervals for PD estimates obtained with open pollination (OPEN) and hand pollen supplementation (SUP) treatments ($x^2 = 7.375$, $p = 0.007$). Different letters indicate significant differences at $p < 0.05$. (c) Scatterplot of PD values obtained through SUP treatment (*y*-axis) in relation to that obtained through NAT (*x*-axis); PD values where PD-SUP>PD-OPEN represented as green dots, PD-SUP<PD-OPEN represented as yellow dots and PD-SUP=PD-OPEN represented as black dots.

3.2 | Methodological considerations regarding hand pollen supplementation

No significant differences were found in PD values among pollen supplementation techniques ($x^2 = 4.106$, $p = 0.250$; Figure S1b; Table S5). However, signs of resource allocation were observed, with significant differences in PD values among experimental scales used in pollination experiments ($x^2 = 8.011$, $p = 0.046$; Table S5). Despite these signs, no significant differences were observed among scales in post hoc tests (Figure S1c; Table S6). Similar results were obtained when rerunning analyses without apple studies (Tables S8–S10).

3.3 | Crop pollinator dependence values

Mean PD values are provided for 141 animal pollinated crops. A list of taxa with PD estimated values is given in Supporting Information (Table S2). Information on specific PD values of crop accessions (including cultivars, varieties, and other infraspecific taxonomic levels) is provided for 94 crops, comprising 317 individual crop accessions (Table S3).

The mean value of PD (PD-final) across all crops of the list was 0.63 ± 0.30 (mean ± SD). The values varied, as expected, from no PD (value of 0) to complete PD (value of 1); however, a concentration of values around 1 was observed, with 26% of the crops having high PD values (PD ≥ 0.90; Figure 3a).

When considering the animal pollinator-dependent classes defined by Klein et al. (2007), 73.8% of the crops were classified as "high" (68 crops, 48.2%) or "essential" (36 crops, 25.5%) (Figure 3a,b), representing a higher number of crops than in Klein et al. (2007). A similar number of crops were observed in the "modest" class in both studies, representing 20.6% of the total crops (29 crops). On the contrary, the number of crops classified as "little" was lower than in Klein et al. (2007), comprising only 5.7% of the crops in our compilation (8 crops; Figure 3a,b).

4 | DISCUSSION

4.1 | Crop pollinator dependence values

This study provides a new compilation of PD values for animal pollinated crops. For several crop species, PD values given here differ from previous global assessments (Klein et al., 2007), with many crops having higher PD values than listed previously. 74% of the animal pollinated crops were categorized in PD classes "high" or "essential", an increased ratio compared to compilations such as Klein et al. (2007). Additionally, compared with previous approaches, the list comprises, for the first time, continuous PD values for 141 worldwide crops, including 317 crop accessions, estimates for 37 crops (highlighted in bold, Table S2) not listed previously or with no data in former global assessments, and detailed data for several crops that were once merged in large groups (see Figure S2).

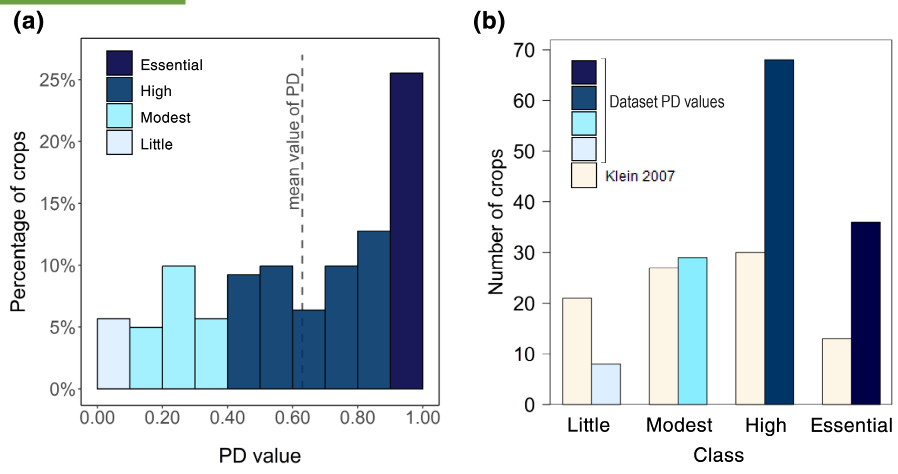

FIGURE 3 (a) Percentage of crops along PD values (0.10 interval range). Final PD was used for each crop (values given in Table S2). Overall mean PD is indicated through a dashed line. Different colour bars represent classes as defined by Klein et al. (2007); (b) number of crops on each PD class: "little" (PD between 0.01–0.09), "modest" (0.10–0.39), "high" (0.40–0.89) and "essential" (0.90–1.00). Beige bars represent the crop's distribution among classes (Klein et al., 2007), and different blue bars represent crops' distribution in this study. Class "no increase" from Klein et al. (2007) was excluded due to the study focus on animal pollinated crops, preventing comparisons in this class.

By providing discriminated PD values for individual crop species and accessions, our study contributes with vital and, until now, neglected information.

Several PD values of individual crops were higher than in previous compilations (e.g. *Citrus*, durian, strawberry, sunflower). These differences are mainly explained by the fact that PD values were obtained through a different methodology, here using hand pollen supplementation instead of open pollination (the primary treatment used in previous estimates) to obtain the final PD value. As hand pollen supplementation accounts for the effects of PL, it provides more accurate PD measures. Once PD estimates are usually based on open levels of pollination, previous studies and compilations are substantially underestimating the importance of animal pollination for crop production.

We found a wide variation in the PD values reported within crops. This might be expected since the degree of self-compatibility and auto-pollination ability has been shown to vary among crop accessions (e.g. sunflower, Carvalheiro et al., 2011; oilseed rape, Hudewenz et al., 2014). Knowledge of the pollination requirements of crop accessions is crucial for suitable management decisions (Hudewenz et al., 2014) and is becoming particularly useful in regions where pollinator loss is, or is anticipated to be, more pronounced (Potts, Ngo, et al., 2016). For example, in pollinator-impoverished locations, when pollinator communities are insufficient to provide the needed pollination services to a crop, selecting accessions that are less dependent on animal pollination may be a suitable solution to ameliorate PL. Unfortunately, 29% of the studies analysed here did not provide information about crop accessions (or any other infraspecific taxonomic level, such as cultivar, variety, forma or clone), hindering the compilation of precise data. Considering the importance of this information (Hudewenz et al., 2014), we recommend that future works should always provide information and data for each accession of the crop under study.

The optimal pollination level from the plant perspective (i.e. plant fitness) differs from that of the farmers perspective (i.e. agronomic and economic yield). To follow farmers' perspective, the PD value was calculated using different production variables, depending on the part of the crop economically used (fruit or seed). Quantity (e.g. fruit set) and quality (e.g. fruit weight) production traits were considered, to accurately account for the impact of animal pollination at both levels. Studies on PD often focus on quantitative variables, with mixed responses between these and qualitative variables (e.g. Bartomeus et al., 2014; Stein et al., 2017). Here, however, only 30% of the entries presented quantity and quality variables. Hence, we recommend that future experiments evaluate production variables related to both levels.

4.2 | Open pollination versus hand pollen supplementation to calculate PD values

Hand pollen supplementation led to higher PD values than open pollination in 50.8% of comparable data points, supporting our predictions. This indicates that PL is common, reducing yield level and, consequently, underestimating potential pollinator's contribution. Therefore, in locations where pollination services are inadequate and/or impoverished, such as landscapes of poor quality due to high levels of fragmentation and/or simplification (Aizen & Feinsinger, 2003; Nicholson et al., 2017), hand pollen supplementation is a more suitable treatment to achieve optimal crop yield and obtain an accurate estimate of PD value. However, despite the importance of accurate PD estimates to value pollinator contribution to production systems, and even though hand pollen supplementation is widely used to study PL in wild plants (e.g. Bennett et al., 2018; Knight et al., 2005), in crops, its use for the calculation of PD has been rare (but see Bishop & Nakagawa, 2021; Garibaldi

BOX 1 Guidelines for pollination experiments when studying animal pollination contribution.

An experimental design should include the following treatments:

a. *Pollinator exclusion*: A bagged treatment, without biotic visits. In crops also pollinated by wind, the experimental design should also evaluate its contribution using two bagging treatments, one using a mesh fabric that allows wind contribution, excluding only biotic interactions, and another using a mesh that restrains pollen movement by both wind and biotic agents. Wind contribution is given by the difference between the two bagged treatments.
b. *Open pollination*: A treatment without any manipulation of the reproductive units where flowers are naturally pollinated.
c. *Optimal pollination* (*or pollen supplementation*): A treatment where flowers are naturally pollinated and to which a hand pollen supplementation is provided. Pollen applications should be performed once or multiple times, depending on the crop's requirements. The use of compatible pollen is crucial, and several sources of compatible pollen should be applied.

Additional notes:

- Bigger scales are preferred (i.e. branch or plant scales).
- Hand pollen supplementations without additional treatments, as bagging or emasculation, are advised but, if additional treatments are essential for the experiment, they can be considered.
- All relevant details should be provided (e.g. accession, cultivar), additionally to details surrounding agricultural management (e.g. application of reproductive hormones, presence of managed pollinators).

et al., 2011; Garratt et al., 2021). Based on these results, we recommend that hand pollen supplementation is included in pollination experiments that aim to assess the contribution of animal pollination to crops. A complete experimental design for such purposes is provided in Box 1.

4.3 | Methodological guidelines for hand pollen supplementations

When performing hand pollen supplementations, assuring efficiency is critical (see Box 1). However, in plant families with complex flower structures or flowers sensitive to manipulation, this can be challenging to achieve. In such cases, animal pollinators may perform better at pollinating than hand pollen supplementation by humans since animals are adapted to exploit floral resources. Thus, the fact that hand pollen supplementation produced lower production values in 25.8% of the data points compared with open pollination is not entirely unexpected. It is possible that in these studies, the supplementation of pollen was not ideal or that over-pollination led to reduced yield (Bishop et al., 2020). This may represent a limitation of the dataset used in our study, which can lead to the undervaluation of PD levels. Indeed, technical approaches used in hand pollen supplementation, such as type of supplementation, scale at which pollination experiments are done and pollen source, are known to affect yield in certain crops (e.g. Webber et al., 2020).

Emasculation of flowers prior to hand pollen supplementation and bagging plants after hand pollination are practices often performed on pollination experiments to exclude production associated with auto-pollination and/or avoid undesirable external pollen, respectively (e.g. Chacoff & Aizen, 2007; Kendall et al., 2020). Here, no significant differences were obtained between standard hand pollen supplementation and supplementation with some of the techniques detailed above, indicating that supplementation using these methods provide reliable estimates of PD or at least, estimates comparable to hand pollinations.

PD values are expected to be higher in pollination treatments conducted at smaller scales (e.g. flower level) than at higher ones (e.g. plant level), as resources for fruit/seed development are usually limited and will be preferentially (re)allocated to flowers with higher pollination quality (Webber et al., 2020). Although no significant differences were observed among different scales, higher PD values were obtained in experiments that used flower as a scale, with marginal *p*-values obtained when comparing flower vs. inflorescence scales ($p = 0.091$). Therefore, more research focused on resource allocation occurrence is needed to fully disentangle the impact of lower scales on associated production levels. Meanwhile, studies should indicate the treatment scale and increase the scale whenever possible to avoid resource allocation problems.

Hand pollen supplementation should be included in crop pollination experiments to account for PL, providing a more accurate method to calculate PD values and assess total pollinator's contribution to crop production. However, it should be bear in mind that the inclusion of hand pollen supplementation increases the time and complexity of such experiments, particularly in mass flowering or self-pruning crops (where sample size needs to be significantly increased to compensate for self-pruning losses) or in plants with complex, fragile flower structures (requiring more time for hand pollen supplementations). Therefore, when designing a pollination experiment, all factors linked with crop reproductive traits should be considered (Young & Young, 1992), acknowledging the limitations and advantages of selected treatments (see Box 1).

5 | CONCLUSIONS

This compilation offers invaluable PD values at crop and accession levels, enabling precise economic assessments of pollinator contribution for individual crop production and supporting the need for pollinator-friendly management in agroecosystems with animal pollinated crops. Our results highlight that the commonly applied method of assessing PD (comparing fruit set in plants exposed vs. isolated from pollinators) can lead to underestimating PD values. Due to this, the value of animal pollination to the production of crops may be higher than the values established in previous studies. Given that most published studies on pollinator's contribution to crops use PD values obtained through methodologies that did not account for pollen limitation, it is probable that pollinator's contribution to crops' local and global production, international trade markets and economic value of pollinators are substantially undervalued. Thus, we recommend that future crop pollination studies consider hand pollen supplementations and include crop accessions when determining PD values for crops. This approach will provide an accurate assessment of the contribution of animal pollination to crop production.

AUTHOR CONTRIBUTIONS

Catarina Siopa, Luísa G. Carvalheiro and Sílvia Castro developed hypotheses and statistical methods, which were discussed with Helena Castro and João Loureiro. Catarina Siopa, Luísa G. Carvalheiro, Helena Castro and Sílvia Castro performed data validation and analyses. Catarina Siopa wrote the first draft, and all remaining authors edited and commented on earlier versions of the manuscript.

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CONFLICT OF INTEREST STATEMENT

None declared.

DATA AVAILABILITY STATEMENT

Additional information can be found online in Supporting Information. Data available in the Figshare Repository at https://doi.org/10.6084/m9.figshare.25350055 (Siopa et al., 2024).

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REFERENCES

Aizen, M. A., & Feinsinger, P. (2003). Bees not to be? Responses of insect pollinator faunas and flower pollination to habitat fragmentation. In *How landscapes change: human disturbance and ecosystem fragmentation in the Americas* (pp. 111–129). Springer. https://doi.org/10.1007/978-3-662-05238-9_7

Aizen, M. A., Garibaldi, L. A., Cunningham, S. A., & Klein, A. M. (2009). How much does agriculture depend on pollinators? Lessons from long-term trends in crop production. *Annals of Botany*, *103*(9), 1579–1588. https://doi.org/10.1093/aob/mcp076

Bartomeus, I., Potts, S. G., Steffan-Dewenter, I., Vaissiere, B. E., Woyciechowski, M., Krewenka, K. M., Tscheulin, T., Roberts, S. P. M., Szentgyörgyi, H., Westphal, C., & Bommarco, R. (2014). Contribution of insect pollinators to crop yield and quality varies with agricultural intensification. *PeerJ*, *2*, e328. https://doi.org/10.7717/peerj.328

Bates, D., Mächler, M., Bolker, B., & Walker, S. (2014). Fitting linear mixed-effects models using lme4. *arXiv* preprint arXiv:1406.5823.

Bennett, J. M., Steets, J. A., Burns, J. H., Durka, W., Vamosi, J. C., Arceo-Gómez, G., Burd, M., Burkle, L. A., Ellis, A. G., Freitas, L., Li, J., Rodger, J. G., Wolowski, M., Xia, J., Ashman, T.-L., & Knight, T. M. (2018). GloPL, a global data base on pollen limitation of plant reproduction. *Scientific Data*, *5*(1), 1–9. https://doi.org/10.1038/sdata.2018.249

Bishop, J., Garratt, M. P. D., & Breeze, T. D. (2020). Yield benefits of additional pollination to faba bean vary with cultivar, scale, yield parameter and experimental method. *Scientific Reports*, *10*(1), 2102. https://doi.org/10.1038/s41598-020-58518-1

Bishop, J., & Nakagawa, S. (2021). Quantifying crop pollinator dependence and its heterogeneity using multi-level meta-analysis. *Journal of Applied Ecology*, *58*(5), 1030–1042. https://doi.org/10.1111/1365-2664.13830

Carvalheiro, L. G., Seymour, C. L., Veldtman, R., & Nicolson, S. W. (2010). Pollination services decline with distance from natural habitat even in biodiversity-rich areas. *Journal of Applied Ecology*, *47*(4), 810–820. https://doi.org/10.1111/j.1365-2664.2010.01829.x

Carvalheiro, L. G., Veldtman, R., Shenkute, A. G., Tesfay, G. B., Pirk, C. W. W., Donaldson, J. S., & Nicolson, S. W. (2011). Natural and within-farmland biodiversity enhances crop productivity. *Ecology Letters*, *14*(3), 251–259. https://doi.org/10.1111/j.1461-0248.2010.01579.x

Chacoff, N. P., & Aizen, M. A. (2007). Pollination requirements of pigmented grapefruit (*Citrus paradisi* Macf.) from Northwestern Argentina. *Crop Science*, *47*(3), 1143–1150.

Dicks, L. V., Breeze, T. D., Ngo, H. T., Senapathi, D., An, J., Aizen, M. A., Basu, P., Buchori, D., Galetto, L., Garibaldi, L. A., Gemmill-Herren, B., Howlett, B. G., Imperatriz-Fonseca, V. L., Johnson, S. D., Kovács-Hostyánszki, A., Kwon, Y. J., Lattorff, H. M. G., Lungharwo, T., Seymour, C. L., & Potts, S. (2021). A global-scale expert assessment of drivers and risks associated with pollinator decline. *Nature Ecology & Evolution*, *5*(10), 1453–1461. https://doi.org/10.2135/cropsci2006.09.0586

Fox, J., & Weisberg, S. (2019). *An R companion to applied regression*. Sage Publications.

Free, J. B. (1993). *Insect pollination of crops* (2nd ed.). Academic Press.

Gallai, N., Salles, J. M., Settele, J., & Vaissière, B. E. (2009). Economic valuation of the vulnerability of world agriculture confronted with pollinator decline. *Ecological Economics*, *68*(3), 810–821. https://doi.org/10.1016/j.ecolecon.2008.06.014

Garibaldi, L. A., Aizen, M. A., Klein, A. M., Cunningham, S. A., & Harder, L. D. (2011). Global growth and stability of agricultural yield decrease with pollinator dependence. *Proceedings of the National Academy of Sciences of the United States of America*, *108*(14), 5909–5914. https://doi.org/10.1073/pnas.1012431108

Garratt, M. P., de Groot, G. A., Albrecht, M., Bosch, J., Breeze, T. D., Fountain, M. T., Klein, A. M., McKerchar, M., Park, M., Paxton, R. J., Potts, S. G., Pufal, G., Rader, R., Senapathi, G. D., Andersson, G. K. S., Bernauer, O. M., Blitzer, E. J., Boreux, V., Campbell, A., … Zhusupbaeva, A. (2021). Opportunities to reduce pollination deficits and address production shortfalls in an important insect-pollinated crop. *Ecological Applications*, *31*(8), e02445. https://doi.org/10.1002/eap.2445

Giannini, T. C., Cordeiro, G. D., Freitas, B. M., Saraiva, A. M., & Imperatriz-Fonseca, V. L. (2015). The dependence of crops for pollinators and the economic value of pollination in Brazil. *Journal of Economic Entomology*, *108*(3), 849–857. https://doi.org/10.1093/jee/tov093

Hedhly, A., Hormaza, J. I., & Herrero, M. (2009). Flower emasculation accelerates ovule degeneration and reduces fruit set in sweet cherry. *Scientia Horticulturae*, *119*(4), 455–457. https://doi.org/10.1016/j.scienta.2008.08.020

Holland, J. M., Douma, J. C., Crowley, L., James, L., Kor, L., Stevenson, D. R., & Smith, B. M. (2017). Semi-natural habitats support biological control, pollination and soil conservation in Europe. A review. *Agronomy for Sustainable Development*, *37*(4), 1–23. https://doi.org/10.1007/s13593-017-0434-x

Hudewenz, A., Pufal, G., Bögeholz, A. L., & Klein, A. M. (2014). Cross-pollination benefits differ among oilseed rape varieties. *The Journal of Agricultural Science*, *152*(5), 770–778. https://doi.org/10.1017/S0021859613000440

Kendall, L. K., Gagic, V., Evans, L. J., Cutting, B. T., Scalzo, J., Hanusch, Y., Jones, J., Rocchetti, M., Sonter, C., Keir, M., & Rader, R. (2020). Self-compatible blueberry cultivars require fewer floral visits to maximize fruit production than a partially self-incompatible cultivar. *Journal of Applied Ecology*, *57*(12), 2454–2462. https://doi.org/10.1111/1365-2664.13751

Klatt, B. K., Holzschuh, A., Westphal, C., Clough, Y., Smit, I., Pawelzik, E., & Tscharntke, T. (2014). Bee pollination improves crop quality, shelf life and commercial value. *Proceedings of the Royal Society B: Biological Sciences*, *281*(1775), 20132440. https://doi.org/10.1098/rspb.2013.2440

Klein, A. M., Steffan-Dewenter, I., & Tscharntke, T. (2003). Fruit set of highland coffee increases with the diversity of pollinating bees. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, *270*(1518), 955–961. https://doi.org/10.1098/rspb.2002.2306

Klein, A. M., Vaissière, B. E., Cane, J. H., Steffan-Dewenter, I., Cunningham, S. A., Kremen, C., & Tscharntke, T. (2007). Importance of pollinators in changing landscapes for world crops. *Proceedings of the Royal Society B: Biological Sciences*, *274*(1608), 303–313. https://doi.org/10.1098/rspb.2006.3721

Knight, T. M., Steets, J. A., Vamosi, J. C., Mazer, S. J., Burd, M., Campbell, D. R., Dudash, M. R., Johnston, M. O., Mitchell, R. J., & Ashman, T.-L. (2005). Pollen limitation of plant reproduction: Pattern and process. *Annual Review of Ecology, Evolution, and Systematics*, *36*, 467–497. https://doi.org/10.1146/annurev.ecolsys.36.102403.115320

Length, R., Singmann, H., Love, J., Buerkner, P., & Herve, M. (2018). Emmeans: Estimated marginal means, aka least-squares means. R package version 1(1), 3.

Licker, R., Johnston, M., Foley, J. A., Barford, C., Kucharik, C. J., Monfreda, C., & Ramankutty, N. (2010). Mind the gap: How do climate and agricultural management explain the 'yield gap' of croplands around the world? *Global Ecology and Biogeography*, *19*(6), 769–782. https://doi.org/10.1111/j.1466-8238.2010.00563.x

Mallinger, R. E., Ternest, J. J., Weaver, S. A., Weaver, J., & Pryer, S. (2021). Importance of insect pollinators for Florida agriculture: A systematic review of the literature. *Florida Entomologist*, *104*(3), 222–229. https://doi.org/10.1653/024.104.0312

Marini, L., Tamburini, G., Petrucco-Toffolo, E., Lindström, S. A., Zanetti, F., Mosca, G., & Bommarco, R. (2015). Crop management modifies the benefits of insect pollination in oilseed rape. *Agriculture, Ecosystems & Environment*, *207*, 61–66. https://doi.org/10.1016/j.agee.2015.03.027

Millard, J., Outhwaite, C. L., Ceaușu, S., Carvalheiro, L. G., da Silva e Silva, F. D., Dicks, L. V., Ollerton, J., & Newbold, T. (2023). Key tropical crops at risk from pollinator loss due to climate change and land use. *Science Advances*, *9*(41). https://doi.org/10.1126/sciadv.adh0756

Mota, L., Hevia, V., Rad, C., Alves, J., Silva, A., González, J. A., Ortega-Marcos, J., Aguado, O., Alcorlo, P., Azcárate, F., Chapinal, L., López, C., Loureiro, J., Marks, E., Siopa, C., Sousa, J. P., & Castro, S. (2022). Flower strips and remnant semi-natural vegetation have different impacts on pollination and productivity of sunflower crops. *Journal of Applied Ecology*, *59*(9), 2386–2397. https://doi.org/10.1111/1365-2664.14241

Nicholson, C. C., Koh, I., Richardson, L. L., Beauchemin, A., & Ricketts, T. H. (2017). Farm and landscape factors interact to affect the supply of pollination services. *Agriculture, Ecosystems & Environment*, *250*, 113–122. https://doi.org/10.1016/j.agee.2017.08.030

Olhnuud, A., Liu, Y., Makowski, D., Tscharntke, T., Westphal, C., Wu, P., Wang, M., & van der Werf, W. (2022). Pollination deficits and contributions of pollinators in apple production: A global meta-analysis. *Journal of Applied Ecology*, *59*(12), 2911–2921. https://doi.org/10.1111/1365-2664.14279

Potts, S. G., Biesmeijer, J. C., Kremen, C., Neumann, P., Schweiger, O., & Kunin, W. E. (2010). Global pollinator declines: Trends, impacts and drivers. *Trends in Ecology & Evolution*, *25*(6), 345–353. https://doi.org/10.1016/j.tree.2010.01.007

Potts, S. G., Imperatriz-Fonseca, V., Ngo, H. T., Aizen, M. A., Biesmeijer, J. C., Breeze, T. D., Dicks, L. V., Garibaldi, L. A., Hill, R., Settele, J., &

Vanbergen, A. (2016). Safeguarding pollinators and their values to human well-being. *Nature*, *540*(7632), 220–229. https://doi.org/10.1038/nature20588

Potts, S. G., Ngo, H. T., Biesmeijer, J. C., Breeze, T. D., Dicks, L. V., Garibaldi, L. A., Hill, R., Settele, J., & Vanbergen, A. (2016). *The assessment report of the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services on pollinators, pollination, and food production* (p. 556). Secretariat of the IPBES.

Power, A. G. (2010). Ecosystem services and agriculture: Tradeoffs and synergies. *Philosophical Transactions of the Royal Society, B: Biological Sciences*, *365*(1554), 2959–2971. https://doi.org/10.1098/rstb.2010.0143

Sáez, A., Aguilar, R., Ashworth, L., Gleiser, G., Morales, C. L., Traveset, A., & Aizen, M. A. (2022). Managed honeybees decrease pollination limitation in self-compatible but not in self-incompatible crops. *Proceedings of the Royal Society B: Biological Sciences*, *289*(1972), 20220086. https://doi.org/10.1098/rspb.2022.0086

Senapathi, D., Goddard, M. A., Kunin, W. E., & Baldock, K. C. (2017). Landscape impacts on pollinator communities in temperate systems: Evidence and knowledge gaps. *Functional Ecology*, *31*(1), 26–37. https://doi.org/10.1111/1365-2435.12809

Silva, F. D. S., Carvalheiro, L. G., Aguirre-Gutiérrez, J., Lucotte, M., Guidoni-Martins, K., & Mertens, F. (2021). Virtual pollination trade uncovers global dependence on biodiversity of developing countries. *Science Advances*, *7*(11), eabe6636. https://doi.org/10.1126/sciadv.abe6636

Siopa, C., Carvalheiro, L. G., Castro, H., Loureiro, J., & Castro, S. (2024). Data from: Animal-pollinated crops and cultivars—A quantitative assessment of pollinator dependence values and evaluation of methodological approaches. *Figshare Repository* https://doi.org/10.6084/m9.figshare.25350055

Siopa, C., Castro, H., Loureiro, J., & Castro, S. (2023). PolLimCrop, a global dataset of pollen limitation in crops. *Scientific Data*, *10*(1), 905. https://doi.org/10.1038/s41597-023-02797-6

Stein, K., Coulibaly, D., Stenchly, K., Goetze, D., Porembski, S., Lindner, A., Konaté, S., & Linsenmair, E. K. (2017). Bee pollination increases yield quantity and quality of cash crops in Burkina Faso, West Africa. *Scientific Reports*, *7*(1), 1–10. https://doi.org/10.1038/s41598-017-17970-2

Tamburini, G., Bommarco, R., Kleijn, D., van der Putten, W. H., & Marini, L. (2019). Pollination contribution to crop yield is often context-dependent: A review of experimental evidence. *Agriculture, Ecosystems & Environment*, *280*, 16–23. https://doi.org/10.1016/j.agee.2019.04.022

Toledo-Hernández, M., Wanger, T. C., & Tscharntke, T. (2017). Neglected pollinators: Can enhanced pollination services improve cocoa yields? A review. *Agriculture, Ecosystems & Environment*, *247*, 137–148. https://doi.org/10.1016/j.agee.2017.05.021

Vaissière, B., Freitas, B. M., & Gemmill-Herren, B. (2011). *Protocol to detect and assess pollination deficits in crops: A handbook for its use* (pp. 1–81). FAO.

Von Liebig, J. (1840). *Die Organische Chemie in ihre Anwendung auf Agricultur und Physiologie*. Vieweg und Sohn. https://doi.org/10.5962/bhl.title.42117

Webber, S. M., Garratt, M. P., Lukac, M., Bailey, A. P., Huxley, T., & Potts, S. G. (2020). Quantifying crop pollinator-dependence and pollination deficits: The effects of experimental scale on yield and quality assessments. *Agriculture, Ecosystems & Environment*, *304*, 107106. https://doi.org/10.1016/j.agee.2020.107106

Wesselingh, R. A. (2007). Pollen limitation meets resource allocation: Towards a comprehensive methodology. *New Phytologist*, *174*(1), 26–37. https://doi.org/10.1111/j.1469-8137.2007.01997.x

Young, H. J., & Young, T. P. (1992). Alternative outcomes of natural and experimental high pollen loads. *Ecology*, *73*(2), 639–647. https://doi.org/10.2307/1940770

Zuur, A. F., Ieno, E. N., Walker, N. J., Saveliev, A. A., & Smith, G. M. (2009). Mixed effects modelling for nested data. *Mixed Effects Models and Extensions in Ecology with R*, 101–142. https://doi.org/10.1007/978-0-387-87458-6_5

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

Figure S1. Difference between PD values for the studied variables.

Figure S2. Values of PD for each crop within *Citrus* and *Prunus* group.

Table S1. Information given in the dataset per entry.

Table S2. Pollinator dependence values of crops—compilation list.

Table S3. Pollinator dependence values of accessions for each crop—compilation list.

Table S4. Number of studies in each form of publication.

Table S5. ANOVA results 1.

Table S6. ANOVA results 2.

Table S7. ANOVA results 3.

Table S8. ANOVA results 4.

Table S9. ANOVA results 5.

Table S10. ANOVA results 6.

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