



Self-Pollinating Crop Isolation Techniques for Micro Scale Gardeners with Limited Access to Arable Land. A Mini Review

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DOI: 10.31080/ASNH.2022.06.1050

Received: April 06, 2022

Published: April 21, 2022

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Abstract

Aims and objectives: The aim of this review is to shortlist and present the most popular crop-isolation techniques for micro-scale gardeners. This will be fulfilled through a mini scoping review of existing literature across academic, professional journals, and grey literature databases.

Introduction: Crop isolation is extremely important in seed saving, especially when growing orphan or landrace varieties of plants. Guidance literature on this matter exists, however, it is mainly orientated around crop isolation for large scale entities/farms and gardeners growing easily crossbreeding food crops. Much less attention is given to micro scale gardeners that try to pursue garden crop cultivation. There is hardly any evidence for crop isolation techniques for self-pollinating plants as those do not require much attention due to their self-fertility properties. Nevertheless, micro scale gardeners have limited access to resources like space, and this increases the chances of producing breed untrue seeds, even from self-fertile varieties of garden crops.

Inclusion Criteria: This review included academic and non-academic (grey) literature written on or around the topic of crop isolation, with a focus on self-pollinating plants.

Methods: The literature search was conducted electronically on the following academic data bases: (1) Science Direct, (2) Emerald Insight, (3) ProQuest, (4) PubMed and (5) Google Scholar. Additional search was conducted using an electronic institutional database/library called Summon Search Engine, with access provided on behalf of the University of West London.

Results: The results indicated that distance isolation is the best technique for minimising cross pollination. In self-pollinating plants, the distance should exceed a minimum of 3 meters. Other techniques like mechanical isolation through blossom bags and insect proof mesh also exist, especially for those that can penetrate the enclosed blossom dome and cross contaminate with foreign pollen. Time isolation is rather difficult to achieve, especially for annual garden crops.

Conclusion: The main issues for pollen cross contamination arises from the lack of arable space amongst micro scale gardeners and allotment holders. It would be advisable not to cultivate multiple varieties in one garden, especially if trying to produce breed true seeds. Although, some of the outlined measures could be applied to minimise the risks involved.

Keywords: Self-Pollinating; Crop Isolation; Micro Scale; Arable Land

Abbreviations

NUS: Neglected and Underutilised (Crops); MI: Mechanical Isolation; DI: Distance Isolation; SF: Self Fertilisation; MSG: Micro Scale Gardeners; SP: Self-Pollinating; CP: Cross-Pollinating; LFS: Local Food System

Introduction

The agricultural practices in the developed world have evolved greatly over the past 2 centuries [1]. From small scale and local farms to super-efficient, mega agricultural holdings where food crops are being measured and monitored with satellite imagery and drones hovering over the fields [1,2]. Historically, the efficiency of the sector started to increase around the First World War and then exceeded during the Second World War [1-3]. This was achieved due to numerous technological advancements happening at that period of time, which were often demand driven by the exhaustive military activities of countries as well as food security related issues. For example, the development of the first artificial fertilizer process, pioneered by Haber and Bosh in 1913 and later the modelling of mass selective breeding techniques which were based on Gregor Mendel's theory of genetic inheritance developed a century earlier [3-5]. These have led to the standardisation of the entire agricultural sector, where farmers have switched from decentralised production of local varieties of crops to cultivating breed true and genetically uniform crops for the purpose of efficiency through maximising yields capacity and producing varieties that would withhold diseases, strong weather patterns and feed the armies and populations in less time [1,2,6].

Neglected and underutilised food crops

As a result of those changes, many local varieties of food crops become cost inefficient and have been lost in time [7]. Nowadays, the search for those crops is becoming ever more popular, as the above agricultural practices have proven to be producing nutrient lacking crops, and lead to many environmental issues like the loss of biodiversity [7,8]. The use of the neglected and underutilised (NUS) crops is still minor as they are mainly used in LFS systems in more rural economies in parts of Europe and developing countries however, there is some growth in popularity, especially across organic markets and local villages and small-town events where farmers exhibit their produce and seed-swap [9,10]. Nevertheless, the overall supply chain of food in the developed world is scarce

in these types of products [2-10]. On the other hand, small scale farmers, gardeners and allotment holders seem to be way ahead of the industry in utilising those forgotten crops. These entities often cultivate orphan, heritage, and heirloom varieties of many food crops [7-10]. Sometimes, even leading to the development of their own landraces that get acclimatised to the unique environment of the allotment or back garden, resulting in improved cultivars that can produce better yield and become more resistant to other localised factors [4,11].

In answer to the loss of biodiversity/agrobiodiversity, many NUS seed banks, seed sharing organisations and communities have sprouted around the globe [12]. These often work on a non-profit basis where seeds are donated to farmers in exchange for fresh and vital seeds the following season [12,13]. This process helps to maintain the vigorous seeds libraries and also gives the growers an opportunity to experiment with forgotten varieties and improve their own food and nutrition security [12,13]. One of the challenges encountered by these micro scale gardeners (MSGs) is the aspect of genetic seed purity, especially when trying to save seeds of specific varieties that have the potential to cross-pollinate with other specimens from the same species [11-14]. Collection and utilisation of breed true crops is crucial in maintaining varietal stability, especially when working with old cultivars like the previously mentioned landraces and other orphan crops that are homozygous [15,16]. Having the ability to save one's own seeds can prove extremely useful to the MSGs as this allows them to collect their own landraces that are suited for that specific and unique environment and therefore, cannot be sourced from external entities.

For the purpose of this mini review, the search was focused on the aspect of minimizing cross-pollination in self-pollinating food crops as many of MSGs have limited access to space, resulting in multiple types of crops being grown across a relatively small area, sometimes, different varieties of the same species are cultivated which allows for 'genetic contamination' even across some self-fertile plants, for example, some legumes (*Fabaceae*).

Pollination

Cross-pollination is essentially the process in which the genetic material 'gamete', in the form of pollen, transfers via various means from one flower onto another flower of a different plant from the same species [16-19]. Self-pollination on the other hand,

is the process of geitonogamy in which the gamete is transferred from flower to flower of a genetically identical angiosperm, usually the same one but this could also happen across two clones or in a gymnosperm as microsporangium passes to the ovule of the same plant [16-19]. Many plants have developed mechanisms like protandry and protogyny, heterostyly, dioecy and chemical activated self-incompatibility, all to stop self-pollination and increase genetic diversity by cross-fertilising [16-20].

Simultaneously, self-pollination in angiosperms occurs when the gemmate is passed from the anthers (male parts) to the stigma (female part) of the same flower in the process of autogamy, which also is a technique to prolong species' existence. Autogamy, or in other words, self-fertilisation (meiosis) is an evolutionary mechanism allowing the plant to reproduce on its own without the reliance on other plants, wind, or animals like insects, making it more likely to generate seeds and therefore, ensure survival of the progeny [16-20]. Self-pollination also successfully maintains many genotypes that would otherwise be lost in the gene shuffle of sexual reproduction, hence helping farmers to produce predictable crops (landraces) [16-20]. Self-pollinating plants ensure autogamy via various means, for example, cleistogamy, where the flowers do not open at all, like most *Pisum sativum* L. varieties and the most common opposite is called chasmogamy, for example in common wheat, (*Triticum aestivum* L.), [19-21]. This is particularly interesting as nearly all angiosperms produce both cleistogamous and chasmogamous flowers on the same plant, therefore exposing the stigma to foreign pollen and encouraging cross-pollination, including the self-pollinating plants [16-21]. Other angiosperms commit autogamy during or right after the flower opening, so there is some possibility for cross contamination of pollen from other plants during that moment of unfertilised exposure. Some plants also self-pollinate through the processes of apomixis or parthenocarpy, which do not require fertilisation and lead to seedless fruit, or seeds that do not require fertilisation, although these Processes mainly occur in perennial fruits like the *Musaceae* family (banana), [16-21] (the science behind the above terms is outside the scope of this mini review). These are the most popular edible crop species that reproduce mainly through self-pollination, either flower to flower or through autogamy: wheat, barley, oats, rice, common beans, peas and tomatoes [16-21].

Implications for breed true seed saving

Maintaining breed true plants is extremely crucial when trying to propagate varieties and when aiming for specific, unique results, therefore, limiting the inter varietal cross-pollination is an important factor [22]. This is extremely difficult for cross-pollinating plants as the pollen can be transferred from plant to plant by humans, animals and rainwater - when in close proximity, [16-20]. Insect pollinated plants get exposed to foreign material more, as some bees and other pollinators can travel hundreds of meters between different plant colonies [16-20]. Other crops like maize cross pollinate significantly easier, where wind tends to be the main carrier of gemmate. In this instance, the plants should be separated with a distance of few or several miles in order to produce breed true seeds [16-20]. This often poses great and costly maintenance difficulties to farmers, especially when considering the factor that the farmer often depends on the neighbouring fields, as those might be the source of foreign pollen.

On the other hand, the isolation process is much simpler with self-pollinating crops due to the anatomy as described above. In order to minimise the risks of cross-pollination, the samplings should be planted 3 meters apart at a minimum, which is significantly less than for cross-pollinating crops [16-20]. This reduces the chances of cross-pollination greatly, but some flowers may still get contaminated by insects that could penetrate the enclosed flower petals in search for nectar or the flower heads can get damaged due to strong winds also carrying pollen [19-21]. It is also important to say, that the MSGs often do not have access to vast amounts of land. As a result of that, distancing may not be enough to prevent cross pollination between different varieties. In this paper, we have outlined further techniques, specifically to the micro-scale gardeners to help in maintaining breed true seeds for self-pollinating food crops [16-21].

Arable land statistics for MSGs in the UK

Average size of a UK household garden ranges from 140m² to 250m² and up to 300m² for an 'acceptable' sized allotment. However, the allotment figures are likely to be elevated as many allotments have been divided into 'halves' or 'quarters', all due to high demand from the public for vegetable growing space. For the household garden figures, those MSGs that grow in these fields, do not tend to use the entire area of their garden, as part of it is also dedicated to leisure, storage and therefore, the arable land figure

is also going to be significantly smaller, perhaps halved. It is also important to mention that many households are deprived entirely of outdoor space or have access to a minimal arable land surface, often the size of few meters squared. According to the Office of National Statistics for the UK, these gardens are often found amongst populations living in large city agglomerations [16-21,32].

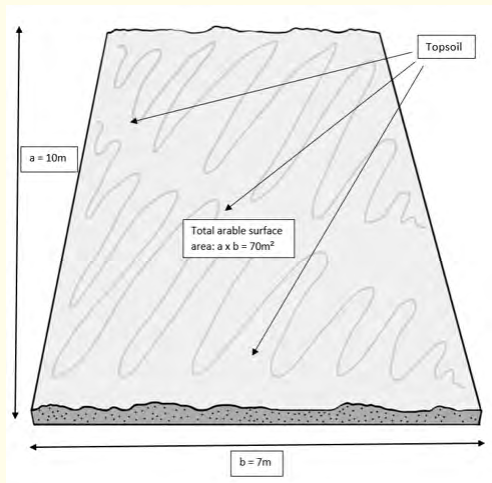


Figure 1: A graphical representation of an average sized arable ‘plot’ of garden, derived from the estimated household garden size in the UK (halved).

Methodology

Search strategy

This mini review was conducted in accordance with the Joanna Briggs Institute guidance for Systematic Reviews for Effective Data Synthesis [23]. The literature search was conducted electronically on the following academic data bases: (1) Science Direct, (2) Emerald Insight, (3) ProQuest, (4) PubMed and (5) Google Scholar. Additional search was conducted using an electronic institutional database/library called Summon Search Engine, with access provided on behalf of the University of West London (UWL, 2022). The following key words were used through the academic data base search: ((self-pollination) OR (crop isolation) OR (crop isolation tech*) OR (breed true seed*) OR (landrace seed*)). The academic search was limited to peer-reviewed and published journal articles, although some non-peer-reviewed material has also been considered for inclusion.

Grey literature was also considered for inclusion. Same keywords were used across the above databases as well as Google, Yahoo and Opera search engines. The searches were not limited by

any date restraints, geographical location of the published articles and the area of study. Articles were although required to be written in English.

Inclusion criteria and shortlisting

The selection criteria for both, peer-reviewed and grey literature was based on topic relevance. Articles that included practical advice on the aspect of crop isolation techniques were considered for inclusion. The first screening stage consisted of an abstract or title page analysis for at least one of the key words, when present, the data piece was taken for further review.

Second data screening was based on full text analysis, where the contexts were analysed for relevance and academic significance. As a result of that, articles that were fully or partially based on the topic of self-fertile crops isolation, were considered for data extraction.

Analysis and evidence presentation

The shortlisted articles were analysed using the NVivo 12 Pro software program, where important parts of text were highlighted and assigned to a specific category, depending on type of isolation (distance, time or mechanical). The data was presented in a written format and supported by technical graphs indicating the garden bed and the localisation of different elements required for the application of the specific crop isolation technique.

Ethics

Ethical approval was not required to conduct this mini review.

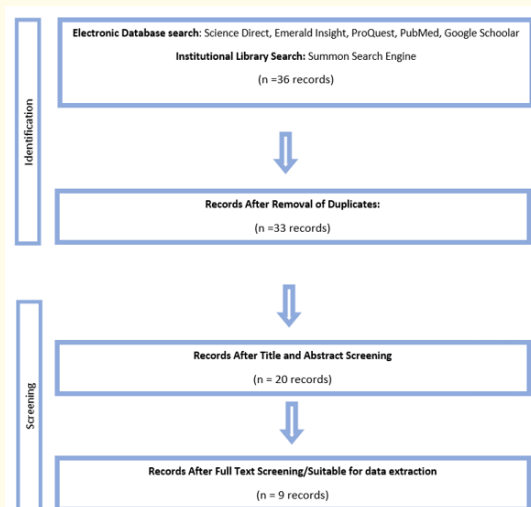


Figure 2: The figure above shows the screening and shortlisting process conducted for the scoping review.

Results and Discussion

The findings for this mini review were based on the primary literature review as evidenced throughout the introduction section, as well as the previously mentioned mini scoping review of academic and non-academic (grey) literature. The overall number of identified references was 31, where 9 were identified through the previously stated methodology, and the remaining were added to the evidence list during the preliminary literature review. The

table below displays the 9 resources in a numerical order, as they appear in the text, including the title of the construct as well as its type. These 9 publications were the prime sources of evidence for the results and discussion section. This section was divided into 4 sub sections, as follows: (1) distance isolation (2) time isolation, (3) mechanical isolation and (4) further techniques to promote self-fertilisation of crops. All of the 9 references included some information on each of the 4 techniques.

No.	Title	Type	Reference
16	Growing Food, A guide to Food Production.	Book	All
24	Simple to complex: Modelling crop pollen-mediated gene flow.	Journal Article (Peer-reviewed)	All
25	Effectiveness of field isolation distance, tillage practice, cultivar type and crop rotations in controlling phoma-stem canker on oilseed rape.	Journal Article (Peer-reviewed)	Distance Isolation
26	Dramatic reduction of crop-to-crop gene flow within a short distance from transgenic rice fields.	Journal Article (Peer-reviewed)	Distance Isolation
27	Isolation distances for Seed Crops: Principles and Practices.	Guide, Academic (non-peer-reviewed)	Distance Isolation
28	Principles and Practices of Isolation Distances for Seed Crops: An Organic Seed Production Manual for Seed Growers in the Mid-Atlantic and Southern US.	Journal Article (Peer-reviewed)	Distance Isolation
29	Isolation Distances, An overview (Seed Savers Exchange).	Guide, Website	Distance Isolation
30	Landrace inventory of the UK. European landraces on-farm conservation, management, and use.	Bulletin, Academic (non-peer-reviewed)	Mechanical Isolation
31	The traditional inexplicable replacement of seed and seed ware of landraces and cultivars: a review.	Journal Article (peer-reviewed)	All

Table 1: Records identified through the scoping review. Please note that the reference column corresponds with the prime focus of the selected article, for example, mechanical isolation or distance and time isolation.

Distance

The literature search has identified 5 articles with a focus on distance isolation (see table 1). Isolation by distance is not a new method for stopping cross pollination, as it has been known for millennia, it was for example practiced by the ancient Greeks [1-5]. When it comes to isolation by distance, the distance itself varies greatly, as there are many dependable aspects that determine the actual distances. The type of cultivated plant being the most important one, whether it is cross-pollinating or self-pollinating (SP), [20,23]. Again, even for self-pollinating flowering plants, the distances can be different, especially when considering large scale

seed production [24-26]. For larger entities, the distance can still be significant for SP crops, especially those that can also get pollinated by insects, the recommended distance by Jeffrey H. McCormack [26] is somewhere along the lines of 15 and 45 meters. Now, talking of micro-scale gardeners that want to save their own seeds, this distance should be no shorter than 3 meters from any individual plant, therefore, a 3 meters radius is required to minimise cross pollination, (see figure 3), especially for crops that tend to produce chasmogamous flowers [24-26]. It would be advisable to plant the different varieties of self-fertile crops in greater distances, for MSGs it could be advisable to plant the crops in the corners of the

garden, but other variables like shade/sunlight exposure and soil depth also need to be considered. It is important to note that these figures are general and other factors like crop varieties, time and physical barriers also need to be considered [26,27]. On that note, it is important to mention that this review has not identified any figures showing the effectiveness of distance isolation, but these metrics are being followed by numerous MSGs as well as mass scale farmers and are believed to be effective [16,24-29].

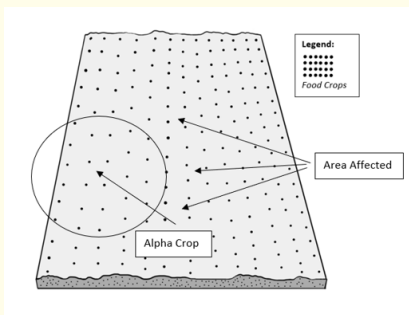


Figure 3: A graphical representation of the 3-meter radius of pollen-contaminated land around the desired (alpha) crop.

Decreasing the minimum distance

The probability of producing breed true seeds from SP plants decreases by around 10% in shorter distances (less than 3m), however there are some methods that can be applied to minimise the impact [16,28]. The articles with a focus on distance isolation (see table 1) have stated that as a general rule, every doubling of isolation distance decreases the amount of cross pollination by a factor of four [16,25-31]. For example, instead of planting the crops in rows the gardener could do it in blocks (see figure 4). This would reduce the periphery significantly, lowering the probability of cross pollination [16,24-26]. Furthermore, only the seeds from the centre orientated plants could be saved as those would be pollen protected by the physical body of the outer layer of the plants. Another mean used to decrease the distance is by selecting seeds produced from flowers that bloomed during the peak moment [25,28]. This technique is often used for open-pollinated plants as the ratio of flowers to pollinators tends to be higher at that particular moment, therefore minimising the chances of contamination by insects. Although, this technique is much more labour intensive as the flowers/seeds would have to be marked individually until the harvest time. If pollinators are the main concern for cross-pollination in self-fertile plants, then the gardener could implement a physical barrier (barrier crop, fence,

mesh) to stop those from coming into physical contact with the plant.

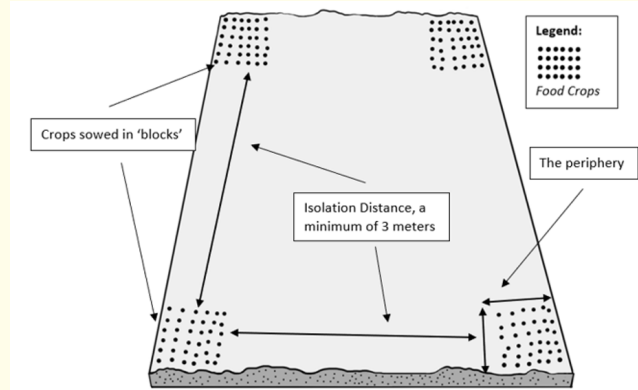


Figure 4: This figure displays the possible block arrangement of crops to increase distance isolation.

Barrier crops

Barrier crops, just like mentioned in the example above (Figure 4), provide a physical barrier for pollinators and can disrupt their flight patterns, especially by affecting the flower constancy behaviours [16,28]. Some pollinators, like bees have a tendency towards being attracted to a singular type of flower and avoid visiting multiple species on single journey from the hive. The incorporation of other angiosperms in-between the desired crops (See figure 5), can help to minimise pollinator exposure by attracting them to the barrier crop, especially when the fluorescent and chemical signatures in the air emitted by the barrier crop are more attractive [16,24,28]. Lastly, it is possible to manually remove the sepals from chasmogamous flowers to make the plant visually less attractive to the insects/pollinators (see figure 6).

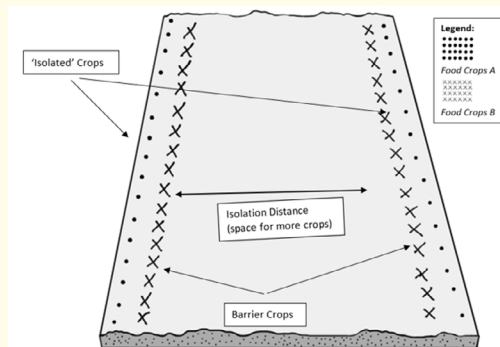


Figure 5: This figure represents the possible incorporation of barrier crops between desired plants to maximise insect distraction.

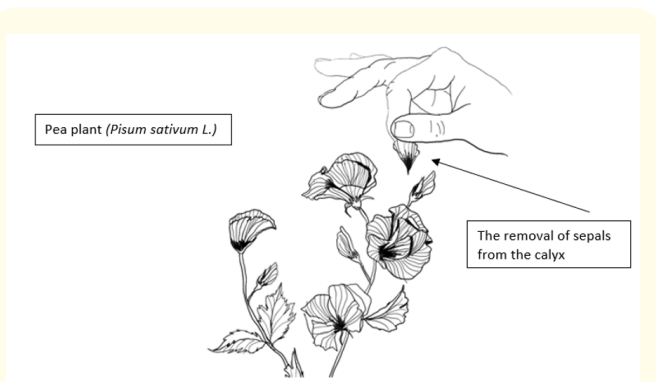


Figure 6: The process of removing outside layer of flowers' sepals/petals (calyx/corolla) to decrease the blossom attractiveness to pollinators.

Time isolation

In theory, time isolation is a relatively easy task, but there are lots of punctuality and mathematical variables. For instance, varieties of plants from the same species tend to have similar flowering periods [30]. Many common beans (*Phaseolus L.*) for example, only tend to bloom once the rest of the plant has achieved maturity and this usually occur 2 months into growth, to minimise the risk of cross-contamination, two different bean varieties could be planted 1 month apart from one another, giving the initial plant more time to mature and develop flowers and fertilise in advance of the second sampling [24-30]. Similar principles could be applied to other self-fertile plants like *Pisum sativum L.* (common peas), although late planting could impact the size and quality of yield, especially when the growth period is extensive and in temperate climates. To conclude, time isolation is not practical for micro scale gardeners, especially when growing annual crops. Efficient time isolation could although work for biennials, two varieties could be planted in 1-year intervals (see figure 7), therefore only one would be flowering at a time [16,25,28,30].

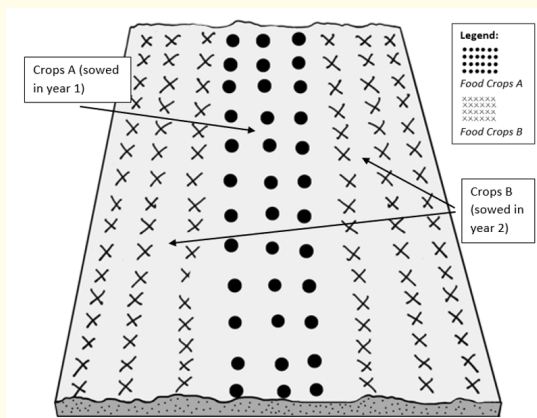


Figure 7: This is a graphical representation of time isolation for biennial crops.

Mechanical isolation

The search strategy led to 1 article with a prime focus on mechanical isolation (MC) and 8 other that mentioned it as secondary in importance, (see table 1). MC is a type of isolation that requires physical barriers in order to be effective [16,25,28]. This is similar to the previously mentioned break crops that act as semi-physical barriers. The barrier should either stop wind pollination by limiting wind movement or by controlling insect pollination by distracting the animals.

MC can take many forms, for example, the crops can be grown in sealed polytunnels or cages. In this instance, foreign pollen will not be able to penetrate the barrier to the blossom and therefore the plant should have enough time to self-pollinate [25,28]. This method probably gives the best assurance of breed true seeds but can be expensive and labour intensive, therefore, it is not recommended for self-pollinating plants but for open pollinated plants instead [27,29]. Nets or insect proof meshes could also be applied, this would work for climbing plants like peas and beans (see figure 8), although this technique is only effective for insects and not wind carried pollen [27,29,31].

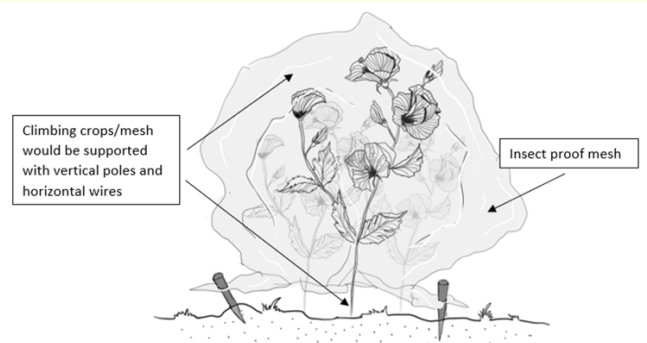


Figure 8: This figure displays the possibility of using insect proof mesh to stop pollinators and other animals like birds from getting into physical contact with the plant. Although the mesh is not going to stop wind carried pollen, it should minimise the already low chances of cross-pollination in self-fertile crops. Furthermore, the mesh is likely to stop larger pests from infesting the plant and the area around it.

Using blossom bags is yet another mechanical isolation technique, in this case, the flower/blossom gets captured and enclosed inside a mini bag that would stop insect and pollen from penetrating (see figure 9). The bag could then be removed after the day of flowering, since the self-pollination usually happens on the first day, or sometimes prior to flowering [16,25,28].

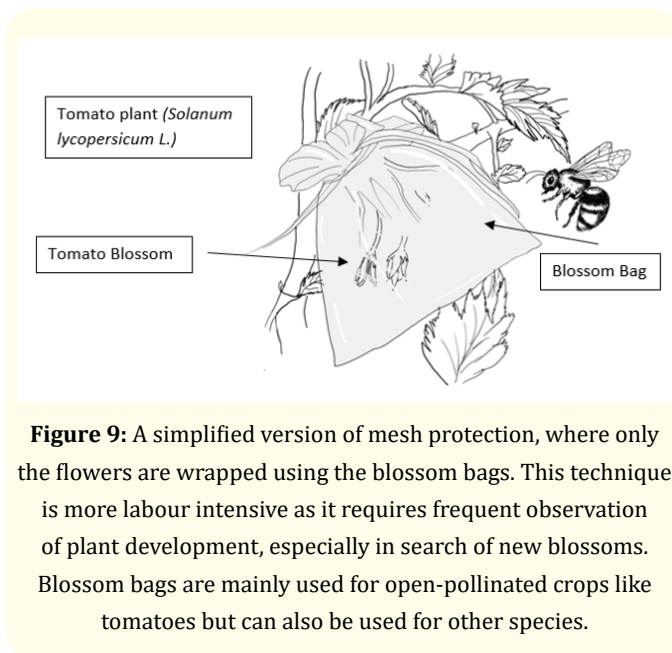


Figure 9: A simplified version of mesh protection, where only the flowers are wrapped using the blossom bags. This technique is more labour intensive as it requires frequent observation of plant development, especially in search of new blossoms. Blossom bags are mainly used for open-pollinated crops like tomatoes but can also be used for other species.

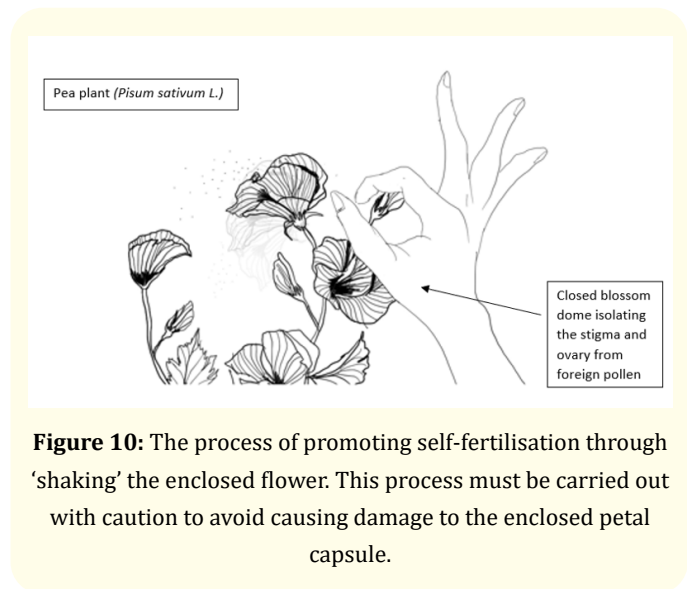


Figure 10: The process of promoting self-fertilisation through 'shaking' the enclosed flower. This process must be carried out with caution to avoid causing damage to the enclosed petal capsule.

Further techniques to promote self-fertilisation

The text above has stated some of the principles of maximising the production of breed true seeds from self-pollinating plants. It is not to be forgotten that the self-pollinating plants make the entire task significantly easier, and investment free [28]. The natural mechanisms present in the plants promote the process of self-pollination and ensure some genetic stability through autogamy [16-20]. Having this in mind, the small-scale gardener could grow multiple varieties of heritage, self-pollinating plants in close proximity and still produce breed true seeds. Precautions are advisable, especially the minimal 3-meter distance but if this is not feasible, the application of other methods should help to isolate the crops [24-31]. Further to that, the gardener could promote self-fertilization itself. The most common method for that, is by touching the flowers and shaking them to help the pollen transfer from the anthers to the stigma in the enclosed blossom capsule (see figure 10). This process is usually carried out in nature by wind and larger animals, but in the allotment or home garden environment, wind might not be strong enough to shake the plant, predominantly due to obstacles like buildings and fences. A useful tool that some allotment holders tend to use is a simple electric toothbrush. This piece of equipment generates micro vibrations, and if applied onto the back of the closed flower, the vibrations should penetrate it and help with fertilisation [27,29,30].

Conclusions and Recommendations

Academic data bases do not contain extensive information on the practical techniques for crop isolation. This is particularly visible for self-fertile food crops as those are believed to be much less technology dependant in producing breed true seeds. Expectedly, there is less advice for micro scale gardeners as that area does not seem to be equally important to researchers and businesses. This is understandable, as the food systems in most developed countries, relay on small, medium and large agricultural enterprises and not on micro gardeners. Nevertheless, more attention is required for the micro scale gardeners, as those tend to grow crops that tangibly elevate the levels of biodiversity. Some of the crop isolation techniques could be used to maximise the probability of getting breed true seeds from self-pollinating plants. The best method is distance separation, but due to reasons discussed previously, other techniques like blossom bags and break crops could also be applied to decrease the probabilities of crossbreeding. Further work should be conducted on the environment and capabilities of micro scale gardeners in growing orphan crops and in developing their own landraces. Also, the potential collaboration between those and various seed banks should be explored further to determine their significance in practical elevation of agrobiodiversity.

Acknowledgements

We would like to thank Klaudia Zborowska for her input into the creation of the technical graphs/images presented in this paper.

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